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CARBONIC ANHYDRASE INHIBITION

III: Effects of Diamox® on Electrolyte Metabolism

THOMAS H. MAREN,[1] BARBARA C. WADSWORTH, ELAINE K. YALE, AND LILLIAN G. ALONSO

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Received for publication July 22, 1954

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INTRODUCTION

The background for the present study has been given in a previous paper on the pharmacology of Diamox® 2-acetylamino-1,3,4-thiadiazole-5-sulfonamide (1). At the onset of this work, the acute renal effects of Diamox (then called 6063) had been thoroughly explored by Berliner, Kennedy and Orloff (2), and the role of renal carbonic anhydrase in acid-base metabolism had been elucidated by Pitts and his colleagues (3). The present study concerns carbonic anhydrase inhibition by Diamox in experimental animals, as it is reflected in electrolyte metabolism for longer periods of time than previously considered, i.e. from one day to over one year.

In a preliminary note (4) we suggested that while the initial renal response to Diamox was largely *independent* of dose, the degree of acidosis induced by continued administration of the drug was directly *dependent* on the magnitude of the dose. Resolution of this apparent paradox provided the basis for our present position regarding the effects of Diamox on acid-base balance. The data on which this depends are given below, and the position may be stated as follows:

A. The *rate* of excretion of HCO_3^- ("buffer base") is unaffected by increasing the dose over that which appears to inhibit renal carbonic anhydrase maximally. Increase of dose, however, *prolongs* the elevated excretion rate of HCO_3^- and increases the acidosis.

B. When Diamox is given intermittently, so that there are suitable drug-free intervals, the animal recovers from the initial loss of bicarbonate. At each ad-

TABLE I

Base Economy in Untreated Dogs

Dog; Wt.	24-Hour Data (9 AM–9 AM), Total Meq.				
	HCO_3^-	T.A.	NH_4^+	T.A. + NH_4^+	Base Economy
M 84, 13.6 kg.	1	10	20	30	29
	7	4	18	22	15
P 20, 12.4 kg.	2	12	35	47	45
	9	11	26	37	28
	5	8	8	16	11
	6	7	19	26	20
M 14, 20.2 kg.	8	8	21	29	21
	5	8	21	29	24
Average	5.4	8.5	21	29.5	24.1
S.D.	±3.1	±2.5	±7.6	±9.4	±10.4

Base Economy = [Titratable Acid (T.A.) + NH_4^+] − HCO_3^-
Animals fed 9 A.M.

ministration of Diamox, the animal is in normal acid-base balance, and renal response is similar to that of the first dose.

C. When Diamox is given on a schedule in which there is little or no drug-free interval, the animal cannot recover from the initial loss of bicarbonate. Depending on the degree of acidosis induced, the renal response to Diamox is then erratic, diminished, or entirely absent.

In this communication, emphasis is laid on the quantitative aspects of acidosis or base depletion. It was found that the effects of various dosage schedules could be most realistically compared by calculating the net renal loss of bicarbonate for each. This was done in either of two ways:

1. For certain short schedules the total renal *output* is available. The base or bicarbonate economy (5) is calculated as NH_4^+ plus titratable acid (T.A.) minus HCO_3^-. From this figure is subtracted the base economy (B.E.) for control periods (Table I), and the net result is loss (or gain) of bicarbonate or buffer base. Specifically, for the experiment of Table V: For the first day the dog received Diamox,

urinary NH_4^+	=	18 meq.
plus		
urinary T.A.	=	8 meq.
minus		
urinary HCO_3^-	=	44 meq.
Base Economy		−18 meq.

For control periods of all dogs (Table I) the base economy averages +24 meq./day. Base depletion is therefore $-18-(+24) = -42$, or a loss of 42 meq., for the first day of drug.

2. A second procedure is applied to the many experiments that lasted for long periods of time, making daily balances impossible. Instead of loss, measurement is made of excess gain of bicarbonate (over the normal of +24 meq./day) observed in recovery periods when drug was removed. Such replenishment of bicarbonate through urinary conservation is assumed (and will be shown) equivalent to the loss maintained while the animal received drug. Thus, in the experiment of Table V, excess $H^+ + NH_4^+$ after the end of treatment was apparent for 2 days. Base economy for this period totalled 80 meq. Subtracting the normal 2-day base economy of +48 meq. (Table I) leaves 32 meq. as the final base depletion for the period of drug intake.

It is of interest to compare base economies as calculated by the initial loss (Procedure 1: 42 meq.) and final conservation (Procedure 2: 32 meq.). The two methods give roughly similar results. The lower figure obtained at withdrawal is to be expected, since inspection of Table V shows that there was some recovery of the initial bicarbonate loss while the dog was on drug. Thus, on day 6, base economy was +29 meq., which is 5 meq. *over* nor-

mal for the colony, or 9 meq. *over* the 3 control days A–C, of Table V. Thus, the gain of 10 meq. (42–32 meq.) from day 1 to the recovery period (7 days) is entirely reasonable.

Although alterations in bicarbonate economy have been used throughout, it is important to show that similar results may be obtained if calculations are based on the excretion of the fixed ions, Na^+, K^+, Cl^-, and $HPO_4^=$.[2] This may be done in a manner comparable to Procedures 1 and 2 above. Thus, for the experiment of Table V, and for the first day (analogous to Procedure 1 above):

3. Urinary sodium + potassium minus (chloride + phosphate) equaled 52 meq. On control days this value averaged 5 meq.; net loss of fixed cations was then 47 meq. and may be compared to loss of 42 meq. calculated by the bicarbonate economy method in 1 above.

4. For the 2 recovery days (analogous to Procedure 2 above) urinary sodium + potassium minus (chloride + phosphate) was −23 meq. Two control days would have yielded +10 meq.; the difference, or 33 meq., represents the extra conservation of fixed cation over fixed anion, and may be judged equivalent to the animal's loss of cation over anion while on drug. This agrees almost exactly with the figure of 32 meq. of bicarbonate, which was conserved at the same time.

Such estimates of total loss or gain of ions have assumed far greater importance in these studies than their concentration in plasma. The former measures acid-base equilibria in the whole body; the latter measures such changes in a portion of the extra-cellular fluid. By measuring the urinary balance of H^+ and HCO_3^- (or of the fixed ions), it has been possible to quantify the distortions of acid-base balance caused by various procedures, and ultimately to relate the diuretic activity of Diamox to the electrolyte pattern of the animal.

Throughout this work emphasis is also put on the relation between the plasma concentration of Diamox and its renal effect. It has been shown (6) that following the first dose of Diamox, or with subsequent doses spaced so

[2] Cooke *et al.* [Am. J. Med., 1954, **17:** 180] have given the important example of recovery from metabolic alkalosis in which a new organic anion appears in the urine. In this case the acid-base equilibrium of the body could not be defined by balance of hydrogen and bicarbonate ions, but was calculated by the urinary changes in fixed ions.

In our experiments no new ion seems to have appeared in important quantities (cf. constancy of cations minus anions in Table V); ammonia production appears adequate in the dog for recovery from metabolic *acidosis*. Nor is there any evidence that tissue production of CO_2 (and hence HCO_3^-) is altered, since initial losses of this substance after Diamox correspond well to its restoration when drug is removed (cf. compare Table IV with Table VII).

In the present studies, therefore, it would seem possible to use *either* urinary balances of $[(NH_4^+ + H^+) - HCO_3^-]$ or $[(Na^+ + K^+) - (Cl^- + HPO_4^=)]$ as a measure of acid-base equilibrium. Specific data to show this are given in the numbered paragraphs 1–4 of the Introduction.

that acid-base balance is preserved, the plasma concentration of drug determines the duration of its renal effect. However, when doses are arranged so that severe acidosis supervenes, the plasma concentration of drug is no longer a direct determinant of urinary composition. We have attempted to document these findings further by presenting the renal and pharmacological data in a unified fashion.

The experimental presentation is divided into three parts: in section A data concerning the primary effects of Diamox on excretion and conservation of HCO_3^- and H^+ are presented; in section B the effects on fixed ions which appear secondary to the principal action of Diamox, i.e. renal loss of bicarbonate ion, are considered; section C is concerned with a number of special factors which have been studied in connection with the renal effects of Diamox.

We have used the term "diuretic" in a broad sense, to cover excess urinary excretion of specified ions, rather than that of water alone.

METHODS

Animal Techniques

The dogs used were beagles of a strain that has been bred and maintained in this laboratory for the past 12 years. Except when noted otherwise, experiments were carried out on females. All experiments were on trained animals without anesthesia. The weights of the dogs are those obtained in the control periods for each experiment. No infusion techniques were used. Some dogs were subjected to a perineotomy, which exposed the urethral opening. In long-term experiments the general procedure was to select certain days for sampling. Apart from these days the dogs lived with other members of the colony in air-conditioned concrete runs. On the sampling or test days they were placed in metabolism cages within these runs. Such dogs usually retained urine completely in the periods under study. *Urine* was collected by catheter into cylinders containing mineral oil or toluene. Spontaneous voidings were collected in bottles containing mineral oil or toluene. The pH of catheterized samples was determined at once. An aliquot was made strongly acid for later determination of ammonia. Bicarbonate and titratable acid were usually analyzed at once; night and weekend samples were run within 12–24 hours. CO_2 was not determined in urines of pH below 6.2. *Venous blood* was collected from the fore- or hind-leg into (a) oiled, heparinized syringes, then expelled under oil into centrifuge tubes, immediately centrifuged at 3000 r.p.m. for 15 minutes and the plasma separated for all determinations except pH; and for pH into (b) heparinized syringes which were then sealed and allowed to equilibrate to room temperature. The pH was determined within 15 minutes of sampling. All stored samples were refrigerated.

Experiments in which arterial (femoral) blood was drawn under similar conditions showed that Diamox did not alter the arteriovenous difference of CO_2 or pH. This difference was the same for 4 control and 4 dogs treated with Diamox, and averaged -1.6 mM CO_2/L. and $+0.08$ pH units in the 8 pairs of samples taken simultaneously from artery and vein. This is equivalent to an A-V difference of -9 mm Hg for pCO_2.

Various feeding schedules were used, and are indicated for the experiments cited. The diet consisted of half Quaker Oats Ken-L-Meal and half horsemeat; each dog usually received 400 g. of the mixture daily. Food analysis was done at intervals through the study; each value is an average of 4 determinations. The Ken-L-Meal contained 9.4 meq. Na^+

and 11.3 meq. K^+/100 g. and the meat contained 2.6 meq. Na^+ and 6.8 meq. K^+/100 g. The mixture as given to each dog per day contained 24 meq. Na^+ and 36 meq. K^+. Water was given *ad lib.* throughout.

Rats were from Carworth Farms, and were albinos of modified Wistar strain. Animals of both sexes were used. For collection of urine, two rats were kept in a cylindrical wire mesh cage over a large funnel which drained into a graduated tube. At the end of the collection period, the bladder was emptied by manual compression. Only sodium and potassium were determined in rat urine.

Chemical Procedures

Analytical methods were conventional unless specially indicated. They are listed below, and apply to both plasma and urine unless otherwise noted.

Sodium and Potassium: Internal Standard (Li) American Cyanamid flame photometer (29).

Carbon Dioxide: Manometric Van Slyke and Cullen. Bicarbonate and pCO_2 from Henderson-Hasselbalch equation using $pK_1 = 6.1$ for carbonic acid (30).

pH: Whole blood: Cambridge meter, with blood microelectrode to nearest 0.01 pH unit. Urine: Beckman Model G meter, open cup, with sealed glass electrode, determined to nearest 0.1 pH unit. 0.014 was subtracted for each degree C below 38°, for both blood and urine.

Ammonia: Urine: Aeration method of Van Slyke and Cullen (30).

Phosphate: Fiske and SubbaRow (31).

Titratable Acid: Urine titrated to pH 7.4 with NaOH using glass electrode.

Chloride: Volhard-Harvey titration (31).

Creatinine: Jaffe method on trichloracetic acid filtrates (32).

Diamox: Maren, Ash and Bailey (7).

Calcium: Plasma: precipitation with oxalate (30) followed by ceric sulfate in place of permanganate titration.

Drug Administration

The chemical and pharmacological properties of Diamox have been described (1). Except where specially noted, Diamox as the free acid was given by capsule to the dogs. Rats received suspensions of Diamox intraperitoneally and subcutaneously, as well as orally by stomach tube. In a few cases dogs and rats received Diamox parenterally as the sodium salt. Such solutions were made by addition of 1.6 moles NaOH per mole of Diamox; pH was approximately 9.1.

Method of Reporting

Each experiment tabulated or graphed is one of at least a pair done on different animals but under similar conditions. The full protocols of the reported experiments and their replicates are available from this Department. Any divergence among these replicates, or any statistical treatment which appears useful, is mentioned in the text.

In the long-term experiments animals were dosed every day according to the schedule indicated, but were studied on selected days shown in the figures or tables.

EXPERIMENTAL: DATA AND COMMENTS

A. Primary Changes in Acid-Base Equilibria

1. Effects of a Single Dose of Diamox

(a) Changes in Urinary Composition

Data. Figure 1 shows the renal effect of a single oral dose of 5 mg./kg. in the dog. The effect of Diamox on urinary composition during the first 6 hours was striking, and may be termed the *drug* phase. Urinary HCO_3^-, Na^+, K^+,

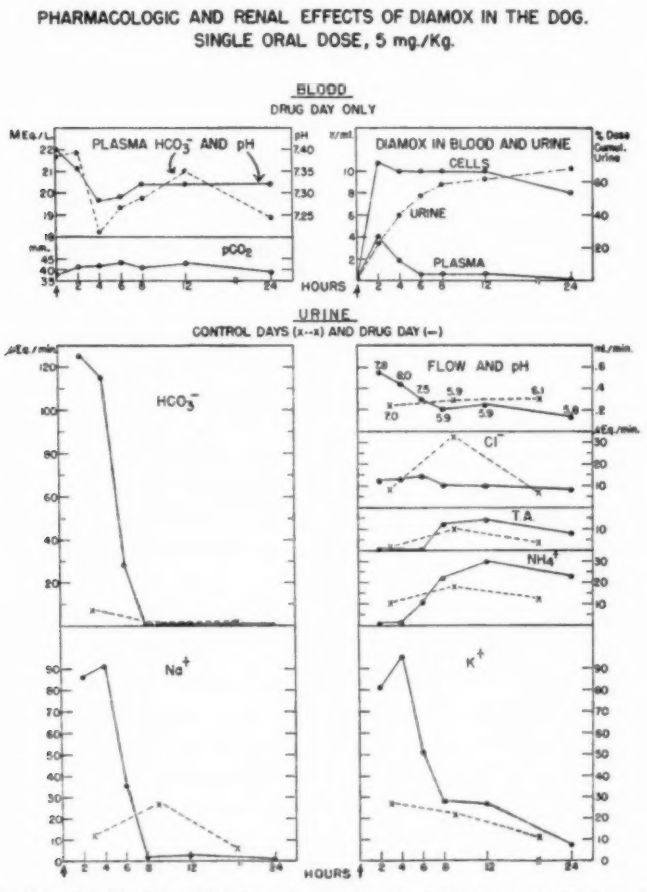

FIG. 1. Dog M 84. Wt. 13.6 kg. 5 mg./kg. Oral. Fed at 9 A.M., which is 0 time. x- - - -x is average of 2 control days, with data plotted at midpoint of collection periods. ●———● is following Diamox.

TABLE IIA

Renal Activity of Diamox in the Rat

Dose mg./kg. and Route	Urine Vol. cc. Hours			Diamox Recovery % of dose, Hours			Excretion of Cations, Meq. Na^+, Hours			Excretion of Cations, Meq. K^+, Hours			Sex
	0–3	3–24	0–24	0–3	3–24	0–24	0–3	3–24	0–24	0–3	3–24	0–24	
0	1	11	12				.14	1.4	1.6	.23	1.5	1.8	m
0	1	9	10				.18	.79	.97	.31	1.1	1.4	m
0	1	15	16				.14	1.7	1.8	.22	2.2	2.4	m
0	2	25	27				.06	.62	.68	.04	1.6	1.6	f
0	1						.05			.09			f
1 i.p.	1	13	14	2	21	23	.13	.79	.92	.09	1.5	1.6	m
1 i.p.	2	14	16	12	15	27	.31	.74	1.1	.13	1.1	1.2	m
4 oral	6			18			.77			.38			f
10 i.p.	5	16	21	15	8	23	.58	.78	1.4	.30	1.2	1.5	m
10 i.p.	4	9	13	24	6	30	.97	.50	1.5	.28	1.0	1.3	m
10 i.p.*	6	12	18	19	9	28	.51	.18	.69	.62	.68	1.3	f
10 i.p.*	5	12	17	40	5	45	.80	.51	1.3	.72	.78	1.5	f
10 oral	8	43	51	19	16	35	1.0	1.3	2.3	.74	1.6	2.4	m
10 oral	7	17	24	19	19	38	.84	.54	1.4	.65	1.1	1.8	m
100 i.p.	4	9	13	12	3	15	.67	.94	1.6	.33	1.2	1.6	m
100 i.p.	5	4	9	4	5	9	.81	.34	1.2	.40	.65	1.0	m
1000 i.p.	7	15	21	4	2	6	1.1	.96	2.1	.63	1.3	1.9	m
1000 i.p.	6	12	18	6	1	7	.9	.95	1.9	.57	1.2	1.8	m
1000 oral	13	27	40			20	1.9	2.0	3.9	1.3	2.7	4.0	m
1000 oral	11	44	55			30	1.5	2.4	3.9	1.0	3.5	4.5	m

Each datum is based on 2 rats per cage. Oral and intraperitoneal drug given in suspension. Animals fed up to time of test, not thereafter. Water *ad lib* throughout.

* As Na^+ salt in solution.

H_2O and pH increased, titratable acid disappeared, NH_4^+ virtually disappeared, and Cl^- was essentially unchanged. Between the 6–8th hour all of these trends (except Cl^-) reversed, with onset of the *recovery* phase. The recovery phase was marked by urine virtually free of HCO_3^- and Na^+, low normal in K^+ and H_2O, and with titratable acid and HN_4^+ well above normal. Reference to the upper right hand diagram of Figure 1 shows that the *drug* phase lasts until the plasma concentration of Diamox falls below 2 micrograms/ml., at which time (6 hours) 52% of the administered drug has appeared in the urine. The persistence of Diamox in the red cells is striking but without apparent effect on the urinary changes. More detailed documentation of pharmacological properties of Diamox are found in Ref. 1.

Comment. The effects on urinary composition illustrated in the drug phase of Figure 1 are near maximal for orally administered Diamox in the dog. This

TABLE IIB

Blood Electrolytes of Rats after 6 Months' Diamox in Diet

Daily Dose mg./kg.	Number of Rats	pH Blood	Calculated pCO_2	Milliequivalents/liter Plasma			
				HCO_3^-	Na^+	K^+	
—	8	7.34–7.54		24–29	122–139	3.5–4.8	Range
		7.43	40	26	130	4.2	Average
100	4	7.29–7.35		19–25	132–136	3.9–4.3	Range
		7.33	43	22	134	4.1	Average
300	4	7.05–7.36		16–23	126–132	3.9–4.5	Range
		7.17	53	19	130	4.1	Average

Animals were sacrificed with pentobarbital.
Blood was taken from the heart before death.
Ref. (1) has details of this experiment. Equal numbers of males and females were used.

may be seen by reference to Figures 5 and 7, which show that for 20 times the dose and 10 times the plasma concentration, the effects on the first 4 hours of Diamox are almost identical to those of Figure 1. Our overall experience with oral Diamox suggests that 5 mg./kg. is a dose which produces maximal effects in about 50% of cases. Ten mg./kg. gives maximal results in almost all experiments. Table IIa shows this type of response in the rat; for the first 3 hours after drug the renal effects are nearly the same within the 100-fold range of 10–1000 mg./kg.

The very high *in vitro* activity of Diamox against carbonic anhydrase (8, 1) together with the small augmentation of effect when plasma concentration is increased above the range of 2–5 γ/ml., suggests that suppression of renal enzyme activity is nearly complete at about this concentration.

The data for the period through which Diamox exerts its diuretic effect on HCO_3^-, Na^+, K^+ and H_2O are in general similar to that of Berliner and his colleagues (2), when allowances are made for the different conditions of the experiments. The sharp reduction, virtually to zero, of urinary ammonia in the drug phase is most significant. Such reduction coincides with the abolition of urinary acidity, and emphasizes again Pitts' concept of the close relationship between renal excretion of acid and ammonia in mammals (3).

The data are compatible with current theories of urinary acidification (3) which regard the rate of H^+ output by the renal tubular cell as dependent on the following reactions:

$$H_2O + CO_2 \underset{\text{Carbonic Anhydrase}}{\rightleftharpoons} H_2CO_3 \rightleftharpoons \underset{\text{Excreted}}{H^+} + \underset{\text{reabsorbed}}{HCO_3^-}$$

Hydrogen ions, added to tubular urine: (a) are exchanged for sodium ions which are then reabsorbed; (b) react with bicarbonate to form carbonic acid,

which diffuses back to the blood as such or after dehydration to CO_2; (c) react with ammonia, which has been split from renal glutamine, to form ammonium ion; (d) diminish tubular excretion of potassium, as if H^+ and K^+ are competing for a common secretory site (2). All of these processes are responsible for the slightly acid character of normal urine.

These characteristics are radically changed by inhibition of carbonic anhydrase, presumably because fewer hydrogen ions per unit time are then available for reaction with lumenal urine. The character of the urine changes in accord with reduction in rate of the four hydrogen-ion-dependent reactions cited just above. With the increase in total electrolyte output, flow also rises.

The *recovery* phase is characterized by excretion of acid and NH_4^+ above normal, and retention of Na^+ and K^+. This is the compensatory response to the acidosis induced by Diamox during the *drug* phase (upper left diagram of Fig. 1). In the low-dose experiment of Figure 1, the recovery phase reached its maximum intensity 6 hours after the end of the drug phase and lasted at least 18 hours. The net effects of *drug* and *recovery* phases in terms of acid-base balance are discussed in the following sections.

(*b*) *Definition of the Acidosis*

A metabolic acidosis is considered due to excess of acid over base in the whole body, as determined by urinary balances discussed in the Introduction. We have considered acid as H^+ (in urine represented by $NH_4^+ + H^+$). Acidosis is net gain in H^+, or net loss of HCO_3^-. In this paper we are concerned with the latter as the primary event, which we term "base deficit" or "base depletion". The measure of this deficit is the measure of acidosis. In metabolic acidosis, plasma pH is below normal, and plasma HCO_3^- concentration is low. However, the present data and current observations (14, 14a) suggest that plasma pH and HCO_3^- concentrations are at best a semi-quantitative indication of the degree of whole body acidosis following treatment with Diamox. Plasma pH may reflect respiratory changes, and plasma HCO_3^- may not represent whole body HCO_3^-. These plasma concentrations do not appear to be primary determinants of the renal adjustment to acidosis induced by Diamox.

A respiratory acidosis is considered due to elevation of pCO_2 in plasma. Compensation refers to adjustment of plasma pH towards normal, in the face of either metabolic or respiratory alterations. In the following sections (i–iii) the effects of Diamox on these various aspects of acidosis are considered.

i. Plasma Metabolic Acidosis

Data. The upper left diagram of Figure 1 shows a drop in plasma bicarbonate and pH, maximal at 4 hours and returning to normal at the peak of the recovery phase. These changes in plasma reflect the trend, but not the quantitative urinary loss of HCO_3^-. The decline in plasma HCO_3^- between 12–24 hours in the absence of renal loss of HCO_3^- must reflect equilibration of this

moiety between plasma and other body compartments during that period. This phenomenon also emphasizes that plasma HCO_3^- concentration is not a reliable measure of base depletion or metabolic acidosis at any one point in time.

Comment. The over-all picture during the first 6 hours represented in Figure 1 is that of a metabolic acidosis following renal loss of bicarbonate and fixed cations, sodium and potassium. It may be noted, however, that pCO_2, which would ordinarily be expected to fall, was instead increased slightly, indicating a respiratory component to the acidosis. This respiratory effect is discussed in the following section.

Instances may be cited where plasma pCO_2 does fall during treatment with Diamox, as in pure metabolic acidosis (9, 10). This is usually under conditions of maximal base depletion (Fig. 11) or of initial high values of pCO_2.[3] Species difference may be involved in the final adjustment of plasma pCO_2 to treatment with Diamox.[4]

That Diamox *can* produce an almost pure metabolic acidosis is shown by days 29 and 30 of the experiment illustrated in Fig. 12. During day 29 the dog lost 60 meq. of HCO_3^- as a result of Diamox administration at 33 mg./kg./8 hours for 24 hours. Plasma HCO_3^- was low and pCO_2 elevated. On cessation of this heavy dosage, plasma HCO_3^- remained low at 14 meq./l. (day 30). At this time little Diamox remained in the body, and pCO_2 fell

[3] Occasionally in the untreated dog when initial pCO_2 is somewhat high, a reduction is observed after Diamox. Such was the case in a duplicate experiment to that of Figure 1, where pCO_2 gradually fell from 50 mm Hg to 40, in the 12-hour period after drug. All other values in the experiment (except, of course, the blood pH itself) were very close to those of Figure 1. This pCO_2 fall was distinctly the exception, but not surprising, in view of the above data indicating the possibilities for change in both directions. The net result could be a rise, fall, or no change. In man (see Footnote 4) the net result is often a fall. It should be emphasized, however, that in all cases after Diamox, plasma pCO_2 is higher than the usual value in metabolic acidosis (under 30 mm Hg after NH_4Cl). In the presence of Diamox there is a constant tendency to keep pCO_2 relatively elevated at the same time that plasma bicarbonate is being lowered.

[4] In man, there appear to be more indications than in the dog, of lowering of pCO_2 on continued treatment with Diamox (15). Acutely there may be no change (13), or a slight drop in some cases (16). The drop in pCO_2 appears to occur both in normals and in patients with elevated pCO_2 due to pulmonary emphysema. The fall in the latter group is usually greater than the fall in the normals. It is important to note that patients with pulmonary emphysema and consequent *respiratory* acidosis as a rule have a good renal response to Diamox (15), hence they tend toward a metabolic acidosis following Diamox.

Similarly, dogs with experimental respiratory acidosis have good renal response to Diamox, even though their elevation in pCO_2 is a stimulus to increased reabsorption of bicarbonate (17, 18, 19). It is significant that the kidney corrects for *metabolic* acidosis by excreting excess acid, in which situation there is impaired response to Diamox (14); in *respiratory* acidosis, however, where the kidney does not make such an adjustment (20), response to Diamox is preserved (15, 17, 19).

precipitously to 28 mm Hg. In this situation withdrawal of Diamox disclosed a partially compensated metabolic acidosis, now uncomplicated by the respiratory effect.

ii. Respiratory Acidosis

Data. The slight upward trend in pCO_2 in Figure 1 has already been indicated. This trend has been seen in most of our experiments and is illustrated in Figures 8, 11 and 12. Although it is slight, it is in the opposite direction to the pattern seen in the metabolic acidosis of ammonium chloride (9, 10) and indicates the presence of a respiratory acidosis. This respiratory component can be isolated by blocking the renal (i.e. the metabolic) effect of Diamox. In the presence of the severe metabolic acidosis following large doses of NH_4Cl, administration of Diamox fails to elicit any renal response but causes pCO_2 to rise sharply with coincident rise in plasma HCO_3^- (14). A similar situation is found in the elasmobranch where Diamox produces no renal effect but appears to interfere with CO_2 transport in the blood-gill system, raising plasma HCO_3^- and pCO_2 (21).

Comment. In the normal dog Diamox produces both a metabolic and respiratory acidosis, with the former predominating except under special conditions. The explanation for the respiratory component is not certain but it might be assigned to inhibition of blood carbonic anhydrase by Diamox. Such inhibition is suggested by our *in vitro* experiments (1), by Tomashefski *et al.* (11), by Shepard *et al.* (12), but not by E. Becker *et al.* (13).

A drop in pCO_2 as a primary effect of Diamox has been seen infrequently.[3] It seems clear that the usual defense (i.e. decline of pCO_2) against lowering of

TABLE III

Base Economy on 5 mg./kg./day of Diamox

	Day	Milliequivalents/Day			Base Economy*
		HCO_3^-	T.A.	NH_4^+	
Control	1	1	10	20	+29
	2	7	4	18	15
Diamox	1	30	11	28	9
	2	22	9	33	20
	3	15	10	33	28
	7	16	8	23	15
	14	16	8	26	18
Average		20	9	29	18
Off Drug	1	2	5	13	16
	2	3	4	9	10

* Base Economy = $NH_4^+ + T.A. - HCO_3^-$.

Dog M 84, 13.6 kg.

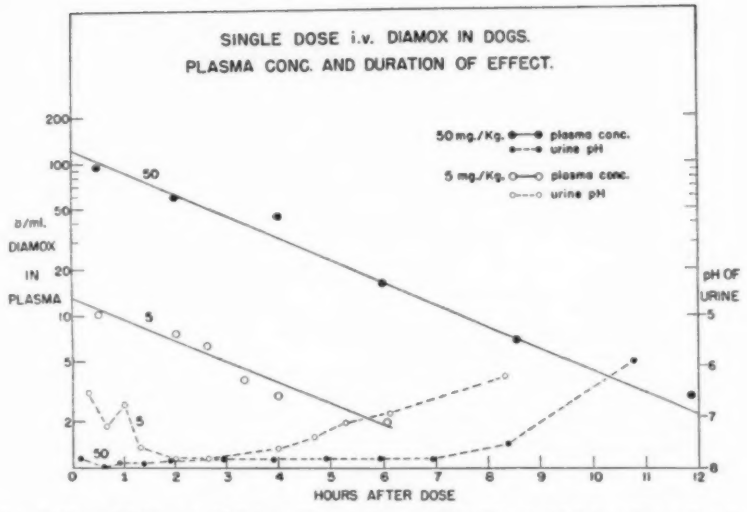

FIG. 2. Dogs M 14, wt. 17.0 kg., 50 mg./kg.; and M 52, wt. 16.9 kg., 5 mg./kg. Intravenous doses. At the higher dose, plasma concentrations are correspondingly higher and more prolonged, and urinary alkalinization lasts longer, than at the lower dose.

pH in metabolic acidosis is not operating fully. The pH is lower after Diamox than after acidifying salts (9, 10) with the same drop in plasma HCO_3^-.

From the data cited under i and ii above, it is evident that depending on the circumstances, Diamox can produce either a rise or a fall in pCO_2. For the conditions of Figure 1 the opposing forces tend to balance so that pCO_2 remains close to normal while HCO_3^- and pH decrease.

iii. Loss of Total Body Bicarbonate

Data. This section deals with the initial daily base or bicarbonate deficit induced by Diamox, which will be expressed as base economy, as defined in the Introduction and as presented for control dogs in Table I.

The experiment of Figure 1 (5 mg./kg./day) is considered in daily base economy terms in Table III. It is evident that on the first day there is a small base deficit. If normal base economy is taken as 24 meq./day (Table I) this deficit is 9–24 = −15 meq., or close to 1 meq./kg.

Base depletions on the first day of treatment with Diamox for all doses are listed in Table IV. (Detailed electrolyte data for these experiments are given below.) It is evident that increase of dose leads to increase of base deficit, owing to prolongation of the effect.

Comment. Increase in size or frequency of dose prolongs the drug phase, which lasts until plasma concentration declines below about 2 micrograms/ml. (See Fig. 2). Onset of recovery is correspondingly delayed, and acidosis, i.e.

TABLE IV

Base Depletion on the First Day of Diamox Treatment*

Dose mg./kg.	No. Tests	Base Depletion meq./kg.	Average	Active† Drug Time, Hrs.
Per day				
5	2	1.0, 1.8	1.4	5 (Fig. 1)
10	2	1.2, 2.0	1.6	~6 (1)
100	4	2.4, 4.1, 5.0, 5.1‡	4.1	15–20 (Fig. 7)
Per 12 hrs.				
10	2	3.4, 3.6	3.5	~12 (1)
Per 8 hrs.				
33	4	4.6, 5.3, 7.5, 8.2	6.4	24 (Fig. 11)

* Calculated in $\frac{\text{meq.}}{\text{kg.}} = \frac{24 - (NH_4^+ + \text{T.A.} - HCO_3^-)}{\text{weight of dog}}$

† Plasma concentration > 2 γ/ml. Data approximated from (1) or given in figures as noted.

‡ Approximate values only: NH_4^+ not determined but calculated as 3 × T.A. This relationship may be observed in Table I (controls) and in subsequent tables for treated animals (Tables III and V). In the tabulation above, base depletion is very largely a function of bicarbonate loss; since reacidification on day 1 is small on high dose schedules, the assumption made here, even if in error by 50%, would not materially affect the final value for base depletion.

base deficit, increased. Thus, although the acute renal effects of Diamox are largely independent of dose, the consequence of increasing a single dose from 10 to 100 mg./kg. is to triple the 24-hour base deficit.

During the drug phase HCO_3^- may be excreted at a rate of 200 microequivalents/min., but during recovery the excretion of NH_4^+ and titratable acid rarely exceed 50 microequivalents/min. Thus, in general, it takes 3–5 times longer in the dog to repair a HCO_3^- or base deficit than to create it. It is evident that sustained base deficit will not occur unless a succeeding dose is given before completion of the previous recovery period. The extent of base depletion, in quantitative terms, will depend inversely on the length of the recovery phase, as judged by drug-free time (24 hours minus active drug time as shown in Table IV).

2. Effects of Single Small Daily Doses of Diamox, 5–10 mg./kg./day.

(a) Dogs Fed on Test Days

Data. Figure 3 shows a continuation of the experiment of Fig. 1 for 2 weeks. Each morning at 9 A.M., about 30 minutes after feeding, the dog received Diamox. Each day there was a response similar to but somewhat smaller than that of day 1. The cycle of 6-hour bicarbonate excretion, followed by extra acidification and NH_4^+ formation, occurred each day. Table III presents the base economy data in daily terms, showing that after the first day, the diuresis of HCO_3^- was matched by excretion of acid and NH_4^+. On day 3 there was a net gain of bicarbonate, offsetting the slight net loss of day 1. As a result, the animal was again in acid-base balance. Plasma pH and bicarbonate

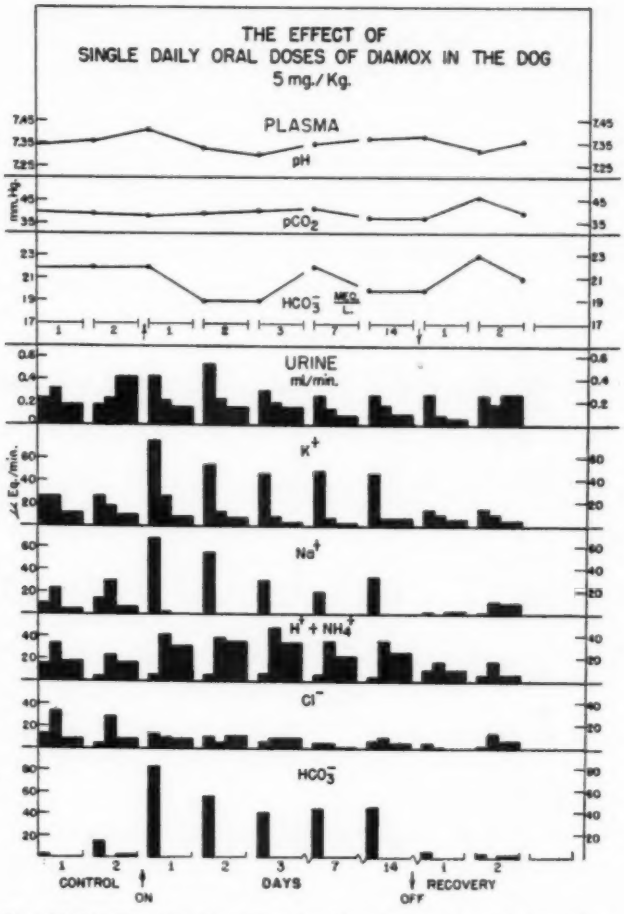

FIG. 3. Dog M 84. Wt. 13.6 kg. 5 mg./kg./day oral on days 1 through 14. Dose and feeding at 9 A.M. each day. Blood drawn at 8:45 A.M. Urine periods 9 A.M.–3 P.M., 3 P.M.–9 P.M., 9 P.M.–9 A.M.

were essentially normal, and on withdrawal of drug there was no change in daily urinary base economy.

Fig. 4 shows a similar experiment at 10 mg./kg./day with the feeding and sampling schedule identical to that of Fig. 3. The cycles of response and recovery are similar to that of Fig. 3, except that the dog on 10 mg./kg./day suffered a slight base deficit during the experiment, as judged by the extra acidification during the recovery period. This excess was 16 meq., indicating a previous deficit of 1.2 meq./kg. It will be noted that this is also the approximate amount of base depletion 24 hours after the *first* dose of 10 mg./kg.

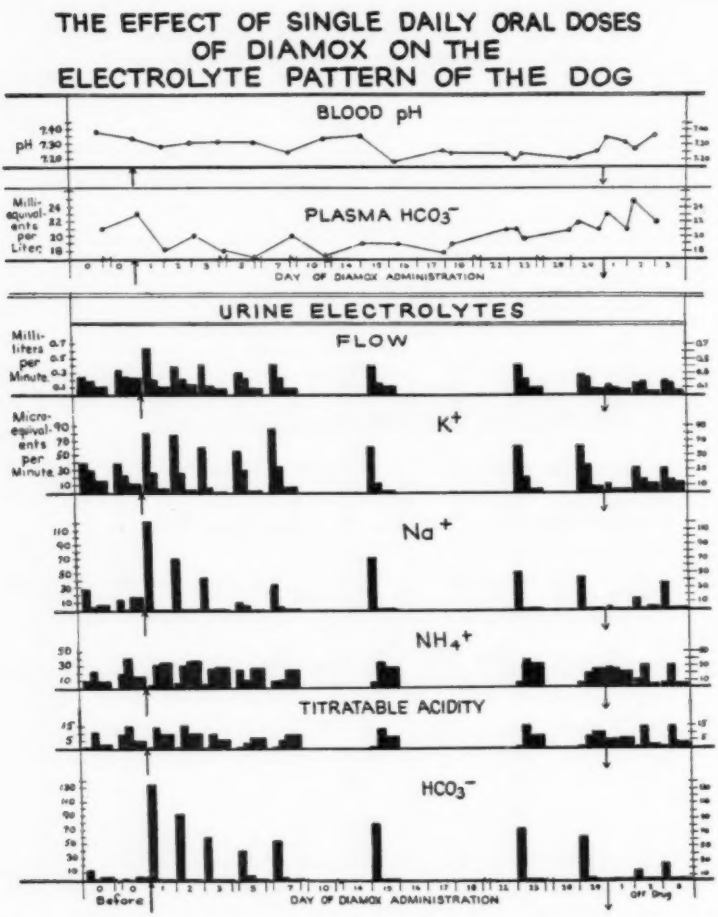

FIG. 4. Dog P 20. Wt. 13.1 kg. 10 mg./kg./day oral on days 1 through 29. Collection and feeding as in Fig. 3.

(Table IV). Correlated with this depletion, HCO_3^- diuresis was less reproducible (for example, day 5 of Fig. 4) than was the case in the experiment of Fig. 3.

Comment. The experiments of Fig. 3 and Fig. 4 suggest that the response to successive doses of Diamox depends on the extent to which the animal has recovered the bicarbonate lost after each previous dose.

(*b*) *Dogs Fasted on Test Days*[5]

[5] Detailed results of these experiments are not given. They are entirely similar to those following similar doses with daily feeding cited above, except that the diuretic effect is less

Data. In dogs fasted on test days after day 1, response (in terms of 6-hour HCO_3^- output) to 5 mg./kg. was approximately half that seen in the fed animal on the same dose (Table VII). At 10 mg./kg. there was a more striking difference between fed and fasted dogs; at exactly the same base deficit, fasting on test days decreased the renal response to one-third that observed in fed dogs (compare A42 and P20 in Table VII). Regular responses in the fasted animal were elicited when 10 mg./kg. was given at 48-hour intervals (P47 in Table VII), where there was *no* base deficit.

Comment. These data suggest that the fasted animal is more sensitive to a base deficit than one fed at the time of drug; the fed animal responds fairly well under a deficit of about 1 meq./kg., but even this small loss changes the response of the fasted animal. In the next section it will be seen that on much higher single daily doses and with a correspondingly high base deficit, there is *no* response on days when the animal is fasted. There is some effect, albeit erratic, when food is given. Finally, it is shown in Section C below (Fig. 16) that both *food* and *time of dose* influence response to Diamox, and that these effects are most pronounced when there is a base deficit.

There is no ready explanation for the prominent effect of feeding on the renal response to Diamox. The amounts of sodium and potassium ingested at each meal (totalling 60 meq.) equal only about 6% of total exchangeable stores of these ions in a 12 kg. dog. This seems too small to be significant. Conceivably, however, this small amount may represent a fraction of fixed cation most readily available for renal excretion under our conditions of dosage with Diamox.

3. *Effects of Single Large Daily Doses, 100 mg./kg./day*

Data. Two dogs (M73 and P27) were placed on this schedule, receiving drug by capsule each morning for 16 months, except for days 141–147 of the experiment. Figures 5–7 give details of the electrolyte and pharmacological data in one of the animals. The dog was fasted on the test days indicated in these figures. In addition, several days were studied on which both animals were fed; these data are included in the summary Table VII.

These dogs were part of our chronic toxicity program and their clinical course was discussed in an earlier paper (1). They showed two significant responses in their electrolyte metabolism, (a) lack of renal response after the first dose, and (b) sustained acidosis.

(a) After the initial renal response there was essentially no further bi-

in the fasted animal after day 1. The fasting situation has the advantage of a lower baseline of control urinary bicarbonate excretion (Table VII), but the disadvantage that consecutive daily data are not possible. For the *first challenge* with Diamox, there is no striking difference between fed and fasted animals (Fig. 16).

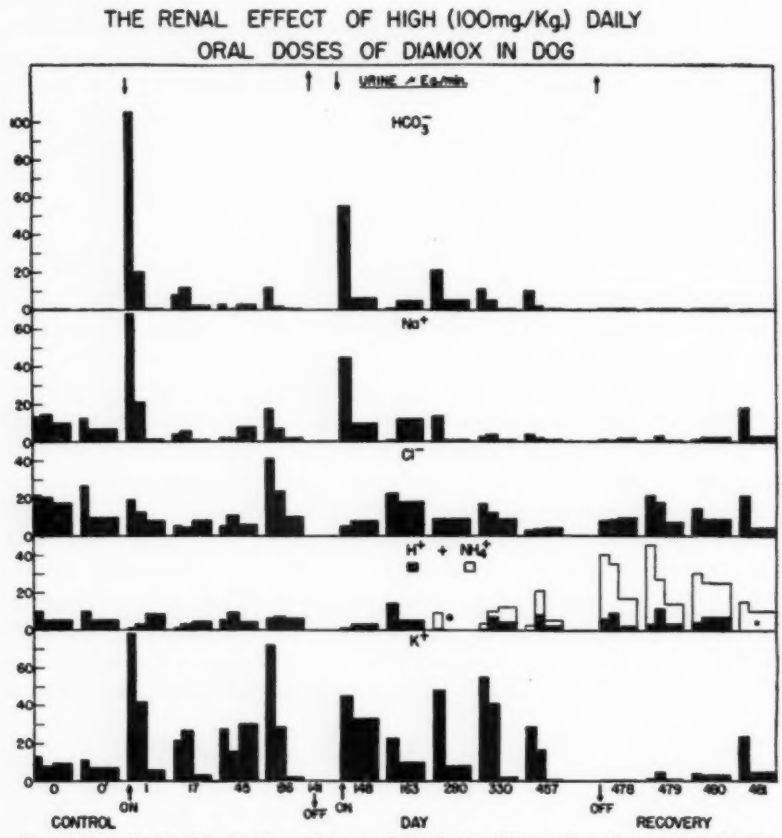

FIG. 5. Dog M 73. Wt. 11.2 kg. Urinary electrolytes. 100 mg./kg./day, at 9 A.M. Fasted on test days. The relative width of the bars indicates the fraction of 24 hours for the sample. ⊕ = H^+ and NH_4^+ not determined; sample contaminated. ★ = H^+ not determined; sample contaminated. Ammonia determinations were not done in the first part of the experiment (through day 163).

carbonate diuresis on this dosage schedule (Fig. 5). When the animal was fed on the test day, response was better, but erratic, from 9–55 microequivalents/min. of HCO_3^- in the 6-hour period after drug (Table VII). The main finding of Fig. 5 is that the urinary pattern virtually reverts to normal after the first few days. On withdrawal of drug for a week, the animals were able to respond again, following which the normal urinary pattern redeveloped.

(b) There was a sustained acidosis, characterized by low blood pH and plasma bicarbonate, elevated plasma chloride, and more or less normal pCO_2 (Fig. 6). The latter component actually fluctuated considerably, both above and below the baseline of 40 mmHg. On the basis both of the first-day deficit

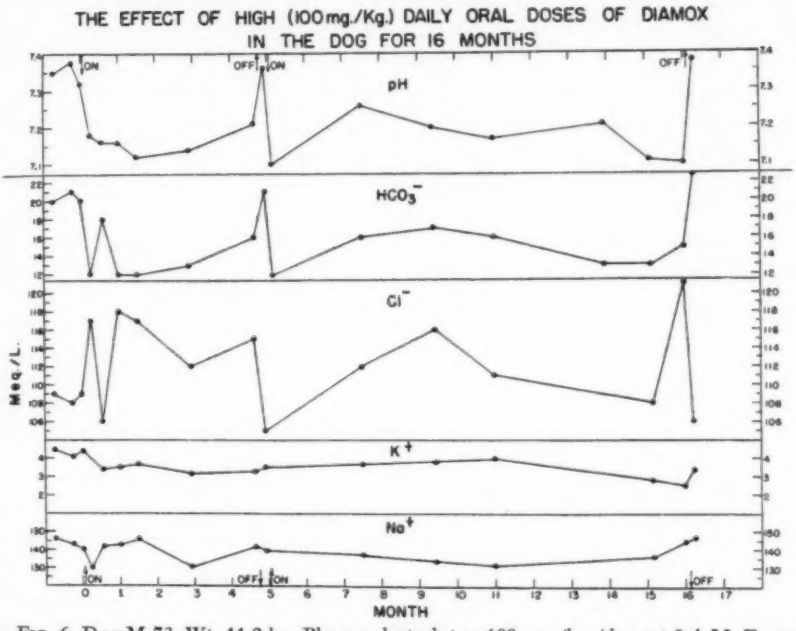

FIG. 6. Dog M 73. Wt. 11.2 kg. Plasma electrolytes. 100 mg./kg./day, at 9 A.M. Fasted on test days. Blood samples taken 24 hours after previous dose. Same experiment as Fig. 5.

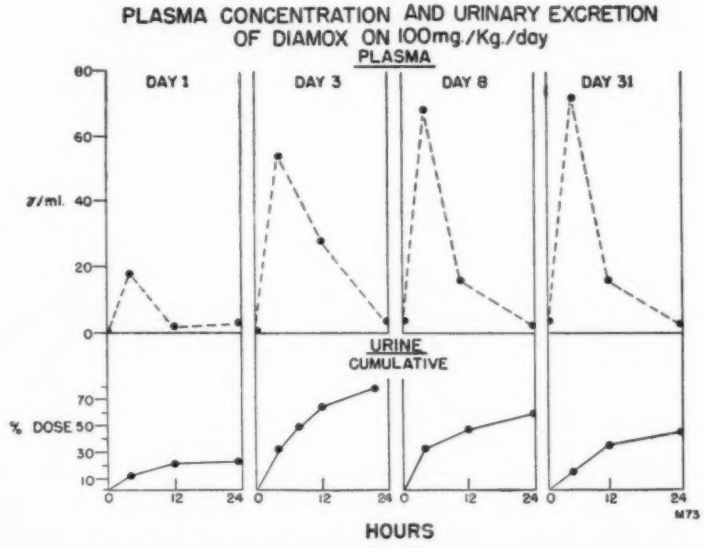

FIG. 7. Dog M 73. Wt. 11.2 kg. 100 mg./kg./day. Plasma concentration and urinary excretion of Diamox, following oral dosage. Same experiment as Fig. 5.

and the extra urinary acidification on withdrawal (day 477), the net depletion of bicarbonate or base deficit was 4 meq./kg. (Tables IV and VII). Urinary and plasma electrolytes reverted to normal in 3 to 4 days following drug withdrawal.

Comment. The two significant results mentioned above, lack of diuretic response and sustained acidosis, are in striking contrast to data obtained with much lower doses. At 5–10 mg./kg. there was just the opposite situation, i.e. continued renal effect and little or no acidosis. The critical consequence of increasing dose is increased *duration* of active level of drug in the body. For 5–10 mg./kg. this is at most 6 hours (Fig. 1) and for 100 mg./kg. it is about 15–20 hours (Fig. 7); Table IV documents this for all doses. Acidosis is the result of increased time during which the drug is present in the plasma in an effective concentration.

The acidosis at the end of day 1 and day 476 for 100 mg./kg./day was essentially the same (compare Tables IV and VII). This suggests simply that the initial base depletion, which is a function of dose and corresponding time of action, was never made up. The normalcy of daily urinary composition after day 1 is consistent with this. Were this urine excessively and continuously alkaline (as day 1), the animals would be rapidly depleted of base; were it excessively acid (as in recovery), the base deficit would be rapidly repaired. The animal is "deadlocked" with a base deficit of about 4 meq./kg. as long as drug is continued and is protected by the acidosis from further depletion. A similar situation is seen in the rat on drug diet (Table IIb).

In the following two sections, the effects of variations in magnitude and spacing of dose are presented. The chief purposes are to find the limits of the induced acidosis, and to explore the relationship between acidosis and lack of renal response to Diamox.

4. Multiple Daily Doses

(*a*) *Moderate (10 mg./kg./12 hours)*

Data. Fig. 8 shows the effect of giving two small doses each day for 2 months. On this schedule there is seen a moderate metabolic and respiratory plasma acidosis. When the dog was given Diamox on fasting days (NF in Fig. 8), there was almost complete absence of HCO_3^- diuresis, while on days of feeding, there was a moderate but erratic diuretic effect. This schedule, which allows about 12 drug-free hours of the 24-hour day (Table IV), clearly does not permit recovery of acid-base balance. The initial base depletion (day 1) was calculated from data of Figure 8 to be 72 meq. On subsequent days during the next 2 months there was no striking loss or gain of base. On drug withdrawal, extra acid and ammonia excretion lasted 3 days, and totalled 56 meq., in fair agreement with the loss of day 1.

Insight into the mechanisms at work in preserving an "acidotic homeostasis"

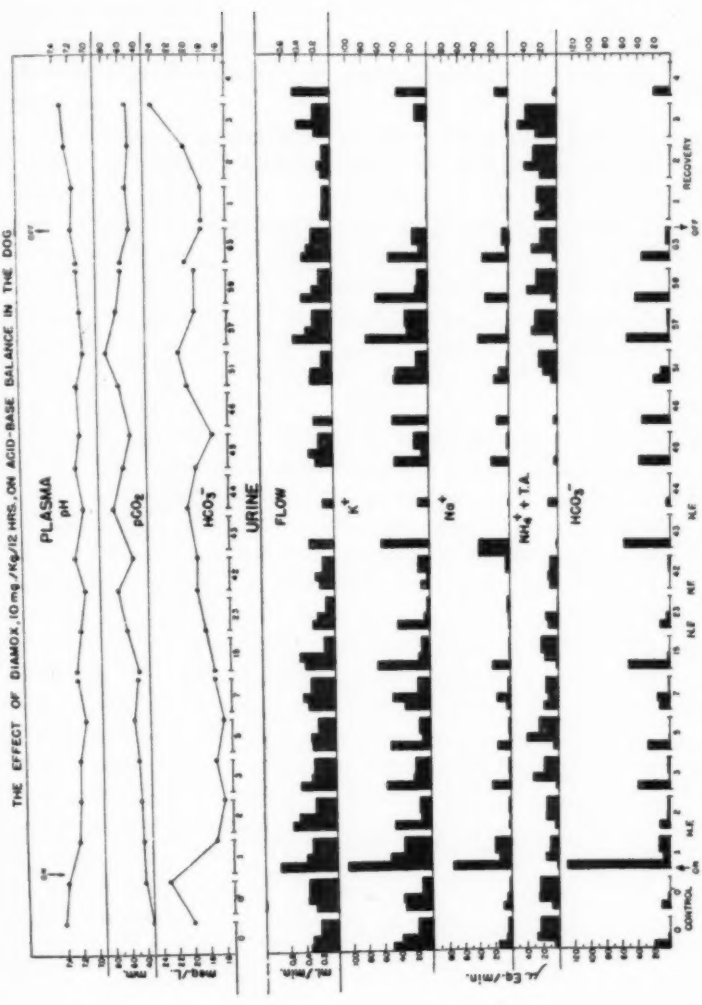

FIG. 8. Dog M 14. Wt. 20.2 kg. 10 mg./kg./12 hours oral. Diamox at 9 A.M. and 9 P.M. Food at 9 A.M. except on days marked NF, where the dog fasted. Blood at 8:45 A.M. Urine periods 9 A.M.–3 P.M., 3 P.M.–9 P.M., 9 P.M.–9 A.M.

is afforded by Figure 9. This shows that on the schedule under consideration, 10 mg./kg./12 hours (B in the figure), bicarbonate loss on day 1 was followed by days in which output of this radical was identical to that of controls (equal slopes of lines). Similarly, after day 1, acid and NH_4^+ output were the same as those of controls. Thus, the initial base loss was neither increased nor repaired. Contrast with one dose per day is of interest (A in Fig. 9). Here the

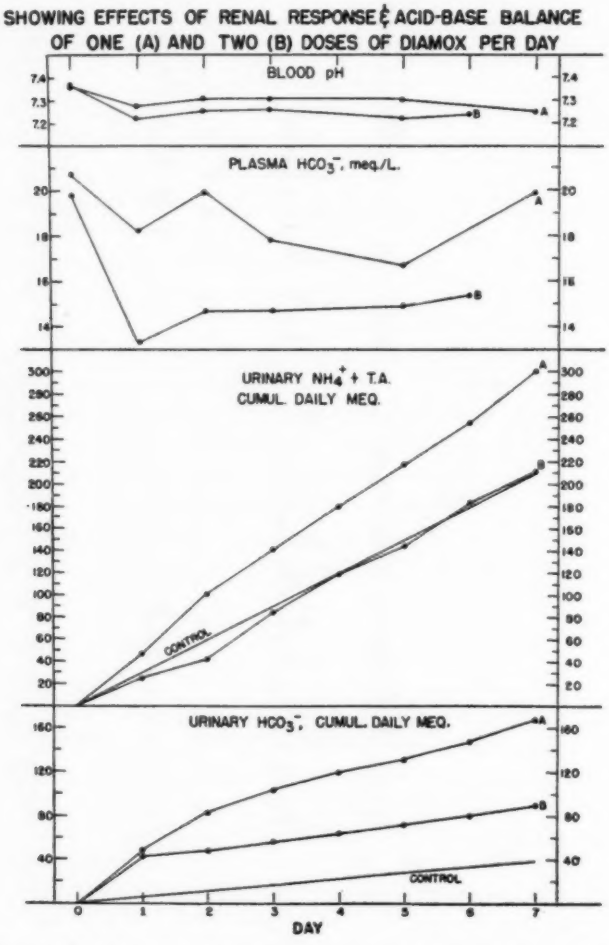

FIG. 9. Dog P 20. Wt. 12.8 kg. Contrasting patterns of electrolytes on 10 mg./kg./24 hours (A) and 10 mg./kg./12 hours (B), in terms of daily output. Control lines are smoothed cumulative values calculated from average output during 4 days without treatment. Blood samples 8:45 A.M. each day. First dose 9 A.M. 0 day.

initial loss of bicarbonate was followed by days when bicarbonate output continued to be strikingly higher than normal (the excess averaging 17 meq./day), as a result of the 6-hour diuresis documented in the preceding sections. Fig. 9 indicates that in this situation excess $NH_4^+ + H^+$ output (averaging 13 meq./day) keeps pace with excess HCO_3^- output. Thus, as has been emphasized above, there is little net loss of base on the single low-dose daily schedule.

Figure 10 shows in more detail how acidosis is prevented on the one-dose daily schedule, and underscores the inevitability of acidosis on the two-dose daily schedule. At night, during the 12–24 hour period after the single daily dose, there was marked augmentation of renal acidification; in the same periods on the two-dose daily schedule, acidification was depressed or at best held at control levels. There were variations around this basic pattern of acidosis and non-response, and for this reason the experiment of Fig. 8 was prolonged for 63 days. It will be seen, for example, that when there *was* a moderate HCO_3^- diuresis in response to the morning dose, there followed extra acidification in the 6–12 hour period (day 58). Urinary composition was again normal in the 12-hour period following the night dose. The net result of this variation is no change in acidosis.

Comment. When two equally spaced doses of Diamox are given each day, acidosis ensues because the dog is prevented, by relatively long exposure to drug, from regeneration of bicarbonate. The base deficit of day 1 persists as long as dosage is continued. There is marked diminution of renal response; on days when the animal is not fed with the morning dose there is virtually no response; when it is fed, the response varies from 10–50% of that of day 1. Following the night dose there is no bicarbonate output but extra urinary acidification is prevented.

This schedule of 10 mg./kg./12 hours allows somewhat more drug-free time per day than the 100 mg./kg./day schedule (See Table IV). We may then expect, and do find, a somewhat lesser acidosis on the former schedule, as judged either by plasma values (compare Fig. 8 and Fig. 6) or by total base depletion (Table VII).

(*b*) *High (33 mg./kg./8 hours)*

Data. Figure 11 shows the results of maintaining a high level of Diamox in the dog for 32 hours. The most significant finding is that after 24 hours urinary composition reverts to that of control periods, even when there are high concentrations of drug in plasma and urine. Here is the converse of the situation depicted in Figure 1; in the present case the pharmacological disposition of Diamox does *not* alone determine the renal effect. In the experiment of Figure 11 the base depletion at 24 hours is 63 meq. and the average 6.4 meq./kg. for four identical experiments (Table IV). Again, the animal is completely deadlocked, and can neither recover (note a low normal, not ex-

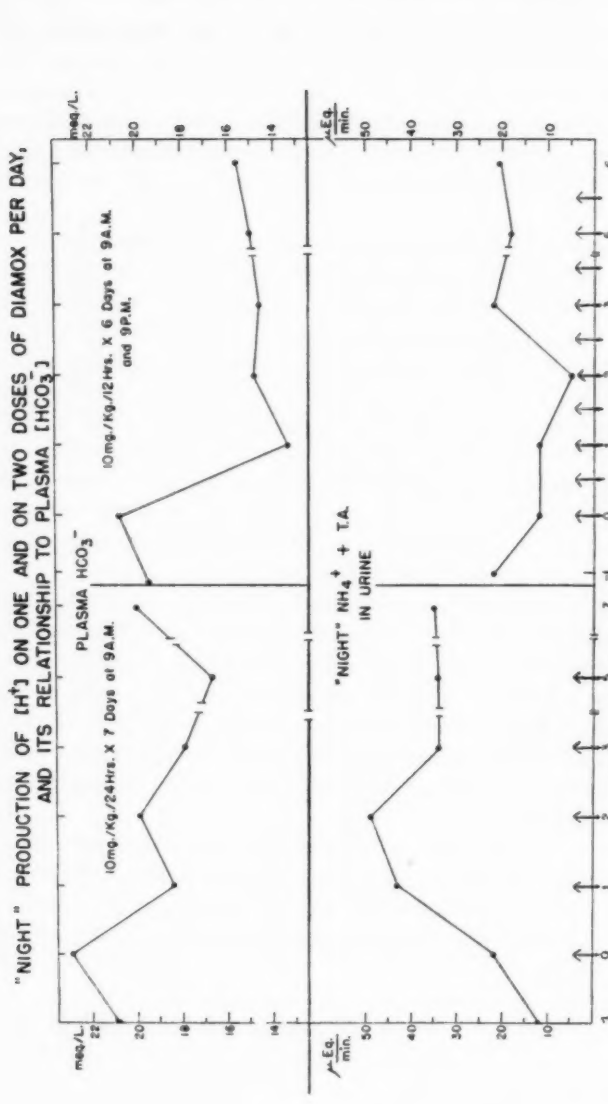
"NIGHT" PRODUCTION OF [H+] ON ONE AND ON TWO DOSES OF DIAMOX PER DAY, AND ITS RELATIONSHIP TO PLASMA [HCO3-]
PLASMA HCO3-
10mg./Kg./24Hrs. X 7 Days at 9A.M.
10mg./Kg./12Hrs. X 6 Days at 9A.M. and 9P.M.
"NIGHT" NH4+ + T.A. IN URINE
mEq/min.
meq/L.
DAY AFTER TREATMENT. DRUG AT ARROWS, 10mg./Kg.

FIG. 10. Dog P 20. Wt. 12.8 kg. Schedules as indicated. Showing augmentation of night acidification after a single small daily dose of Diamox. Blood samples at 9 A.M. just before dose. Fed 9 A.M.–10 A.M. Night urine collected 9 P.M.–9 A.M., and graphed as of 9 A.M. on the day indicated. First dose at 9 A.M. of 0 day.

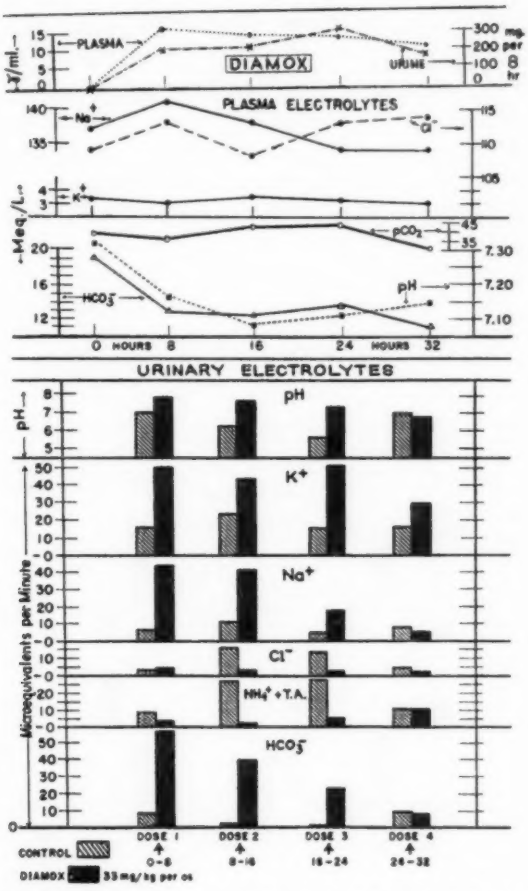

FIG. 11. Dog M 78. Wt. 10.6 kg. 33 mg./kg. every 8 hours x 4 doses. Samples of blood and urine taken every 8 hours for 4 periods. Without drug (hatched); with drug (solid) every 8 hours from 0 time.

cessive $NH_4^+ + H^+$ output) nor lose further HCO_3^-. It continues in the same situation as long as the drug is given (Fig. 12); plasma bicarbonate is between 12–14 meq./l. and pH between 7.1 and 7.2. The plasma pCO_2 shows an initial rise, then a fall, indicating both a respiratory and a metabolic component to the "plasma" acidosis.

The severe acidosis gradually disappears when drug is withdrawn; renewal of dosage (day 29) reveals an excretory and plasma pattern almost identical to that of the first day of dose (Fig. 12). After this renewed dosage, recovery was studied on the following day and showed the highest acidification rate in

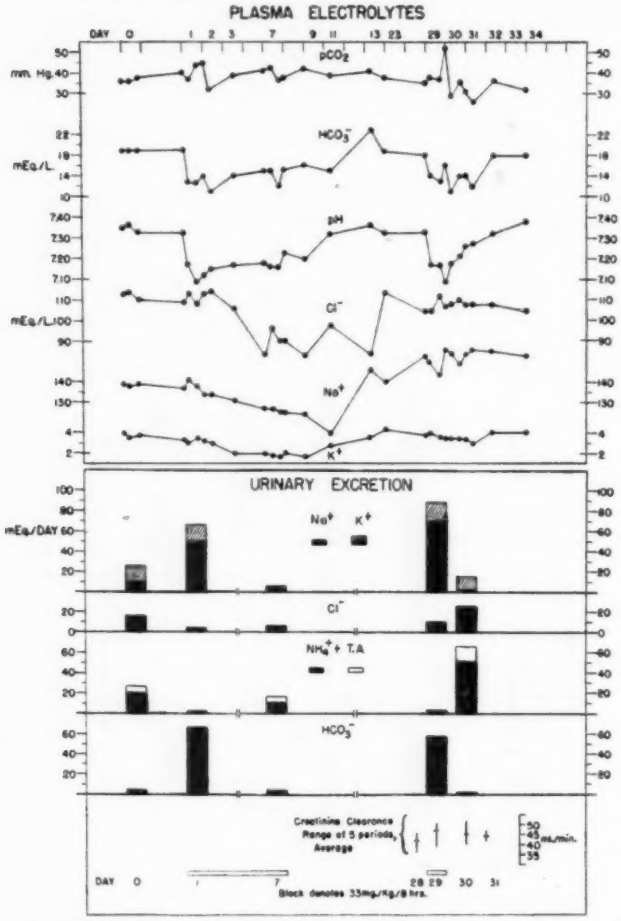

FIG. 12. Dog M 78. Wt. 10.6 kg. 33 mg./kg./8 hours for 7 days, followed by a recovery period and final challenge 2 weeks later. On days 3–9 there was vomiting and no food intake.

our experience, 67 meq./24 hours. The plasma changes were of interest; when drug was withdrawn and before the metabolic base deficit was restored, pCO_2 fell precipitously to 28 mmHg., indicating again that in the presence of the drug, pCO_2 had been elevated.

Comment. A significant sequence in the development of drug tolerance in this experiment is the change in plasma bicarbonate on the one hand, and in total base depletion on the other. It is clear from Figure 11 that after the first 8 hours, plasma bicarbonate reached its minimum value of 13 meq./l.

When Diamox was given at this point there was good bicarbonate diuresis. Sixteen hours later, with no change in plasma bicarbonate, the bicarbonate diuresis was abolished. In this interval, however, net body depletion of HCO_3^- had risen from 26 to 63 meq./kg. This suggests that plasma bicarbonate *per se* does not play a major role in determining renal reactivity to Diamox. Strong support is given to this premise by the experiments cited below on a dog with low plasma bicarbonate, apparently due to hyperventilation (Section C.4).

Continuous exposure to Diamox in the dog thus appears to induce a metabolic acidosis with base depletion of approximately 5–8 meq./kg. (see also Table IV). In this situation Diamox has no diuretic effect and urinary composition is like that during control cycles. Diamox prevents recovery from the acidosis; at this level of base depletion and in the presence of drug, there seems insufficient stimulus for extra urinary acidification (see Discussion). The result is a new acidotic homeostasis. This degree of acidosis (5–8 meq./kg. base deficit) appears maximal for our experience with Diamox.

Continuous exposure to Diamox in the rat (Table IIb) also results in acidosis, the plasma parameters of which indicate both a metabolic and a respiratory component. As in the dog the degree of acidosis depends on dose.

Certain factors other than acidosis which might contribute to development of renal resistance to Diamox appear to be excluded by the following. Hyperchloremia may be ruled out by data in Figure 12 (day 7), which shows complete resistance to Diamox in the face of severe *hypochloremic* acidosis. In this case chloride depletion was due to vomiting. Potassium depletion as a major factor in resistance may be tentatively ruled out by analogy to failure of response to Diamox induced by giving NH_4Cl 1 hour before the drug (14). In this case K^+ loss is negligible.[8] In a variety of situations, therefore, we can show that Diamox becomes less effective in promoting renal base excretion as acidosis increases.

B. Synopsis of Effects on Other Ions

The experiments of Part A also show the effect of Diamox on the balance of sodium, potassium, chloride, and phosphate ions. An attempt will be made here to evaluate their place in the metabolic changes produced by Diamox.

1. Sodium

As the foregoing data show, administration of Diamox to a normal animal results in renal loss of bicarbonate ion which is matched by diuresis of sodium and potassium. It has been shown that the rate of excretion of these ions is independent of dose, but that at very high doses the diuresis is prolonged enough to increase materially the amount of base excreted (Table IV). For sodium, assuming that the dog contains about 40 meq./kg. (22), not more

than 10% of this amount is lost in the various experiments described here. Following an initial loss of sodium, there are no further losses on continued administration of drug. One of two situations obtains, either (*a*) a renal response but no net loss, or (*b*) no renal response: (*a*) On the small dose, where there is a renal response after each daily administration of Diamox, the sodium output appears adjusted so that no *net* loss ensues. Excretion of acid and conservation of sodium in the 18 hours following the diuretic response (Figs. 3 and 4) preserves the balance of this ion, so that on daily administration of drug there is little or no net renal loss (Figs. 13 and 14). It may be seen (Fig. 13) that when food intake is diminished, Na^+ output tends to be reduced,

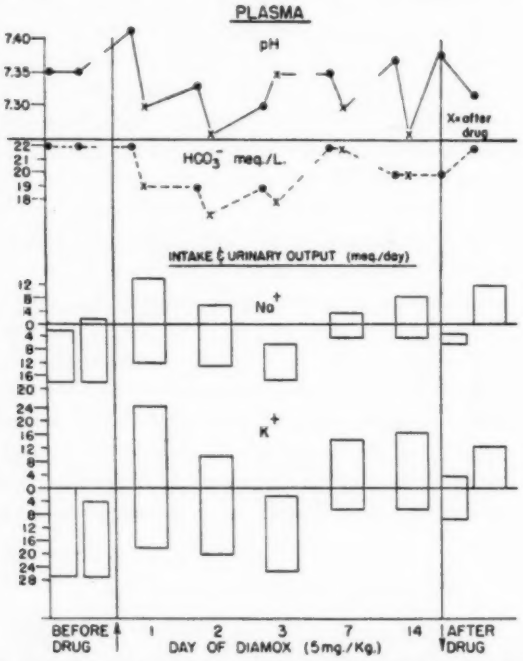

FIG. 13. Sodium and potassium balance in Dog M 84. Wt. 13.6 kg. Oral Diamox 5 mg./kg./day at 9 A.M. Fed 9 A.M. as described under Methods. Balances are plotted as suggested by Reifenstein, Albright and Wells (27). Intake is the distance from the baseline (0) down to the base of each rectangle; urinary output is the total height of the rectangle. Thus when the rectangle is *below* the baseline, balance is *positive*; when it extends *above* the baseline, it is *negative*.

Note that this dog ate erratically. Note tendency for negative base balance when Diamox is given at periods of low food intake.

● represents plasma concentrations at 9 A.M. (drug time); x represents such values 6 hours later.

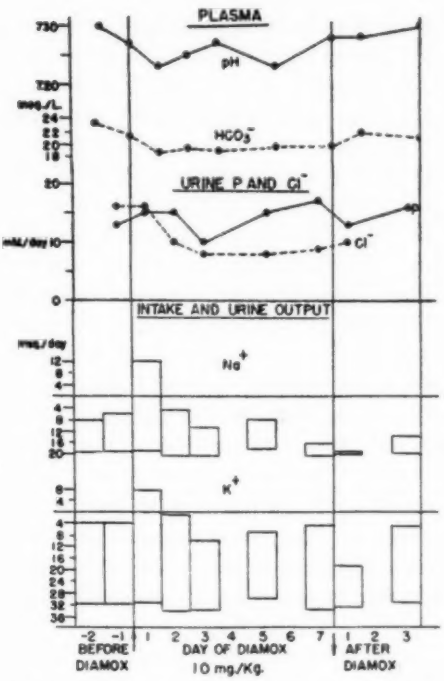

FIG. 14. Sodium and potassium balance and urinary inorganic phosphorus and chloride in Dog A 42. Wt. 14.8 kg. Daily oral Diamox 10 mg./kg./day at 9 A.M. Fed 4 P.M. For explanation of figure see legend for Fig. 13. Note regular food intake, and positive balance of sodium and potassium.

even during drug administration. In Figure 14 it will be seen that Na^+ balance in a dog receiving Diamox can be markedly positive. (*b*) On the large dose or multiple doses, with resulting acidosis (Figs. 5, 8, 11, 12), there is usually no renal response after day 1.

Severe depletion of sodium has not been observed, except when vomiting accompanied high and frequent dosage with Diamox. However, even with this unusual combination of initial sodium loss with bicarbonate and later sodium loss with chloride (Fig. 12), the animal recovered, even though the dosage and the symptoms continued for 5 days. Plasma sodium declined to 115 meq./l. Renal loss of sodium after day 1 was small.

The rat responds typically to Diamox (Table IIa), but withstands very large doses in the diet with resultant acidosis but no evidence of progressive sodium or potassium depletion (Table IIb). Similar considerations appear to apply to man (15).

The degree of sodium depletion may be gauged by its retention on recovery. The largest depletion in the present series (excluding episodes of vomiting, see above) was in the experiment of Figure 5, where, on drug withdrawal, the dog put out a sodium-free urine for 3 days on an intake of 16 meq./day. Depletion can be reckoned as 48 meq., or about 10% of body sodium in a 12-kg. dog. Plasma sodium was lowered by about 10–15 meq./l. reflecting the total deficit.

In the dog, a first dose of oral Diamox usually leads to greater excretion of potassium than sodium. In 26 experiments, the average urinary K^+/Na^+ quotient was 1.3. The converse is true of the rat, where the experiments of Table IIa show this quotient to be 0.7.[6] In man, evidence thus far suggests that in normals the ratio is closer to the rat than to the dog (15, 23a). In retrospect, this is an interesting illustration of the value of using several experimental species; it appears that there is nothing about carbonic anhydrase inhibition generally that demands greater excretion of one over the other major fixed cation.

Finally, when there is abnormal retention or excretion of sodium (or potassium) these observations on the *normal* animal cannot be expected to apply. The edematous cardiac patient on Diamox may lose relatively *more* sodium than the normal dog, or may be entirely refractory even to the first dose (23, 24).

2. Potassium

Berliner and colleagues (2), on the basis of studies with Diamox, postulated that H^+ and K^+ compete for an active excretory mechanism in the renal tubular cell; when the excretion of one of these ions is blocked, that of the other increases. Thus, alkaline urine will generally be rich in potassium. This is the case following Diamox administration, bicarbonate infusion, or hyperventilation alkalosis. Our experience in the present series, therefore, agrees with the concept of Berliner.[7]

The question of whether administration of Diamox is a serious threat to the potassium content of the body has been raised repeatedly. We have indicated in the previous publication (1) that repeated huge (1000 mg./kg.) daily intravenous doses of Diamox for 3 days led to the death of two dogs, and that in the terminal phase both had plasma potassium values under 1.5 meq./l. Only in these extreme circumstances, however, has it been possible to produce lethal depletion of potassium in the experimental animal.

The initially depleted animal has regulatory mechanisms for conservation

[6] The rat has the same K^+/Na^+ ratio in his diet as the dog. The rats on these acute studies were maintained on Purina Dog Chow Checkers, which contain 16 meq. Na^+ and 22 meq. K^+ per 100 g. See Methods Section.

[7] Unexplained by his theory, however, is the high K^+ concentration of extremely acid urines, as in dogs receiving NH_4Cl.

of potassium, just as for sodium. There are, however, some quantitative differences. Initially, in the dog on Diamox, about 12–15% of the total body potassium of 40 meq./kg. may be lost over a 3-day period, whereas with sodium, net loss is somewhat less (see above) and is limited to the first day. Clearly, in this species at least, the brake on potassium excretion does not come into play as soon as that for sodium. It appears that once the animal passes this early critical period, it is in no danger of toxic depletion of potassium, particularly if food intake is not decreased. It must be recognized that these dogs are taking in about 50% more potassium than sodium in the diet, and can afford to lose considerably more K^+ in the initial period.

In the dog, conservatory mechanisms for potassium appear more sluggish than those for sodium. We have referred (1) to a period of poor food intake during which one of the high-dose dogs (M73, see also Figs. 5–7) had low plasma K^+, although there were no untoward sequelae from this episode. Figure 13 indicates 2 days of negative potassium balance associated with poor intake in another dog; in the similar schedule of Fig. 14, however, where intake was regular throughout, there are no periods of negative balance after the first day. Food intake is thus a critical factor in K^+ balance during Diamox treatment. This drug may be a potential threat to K^+ balance, but in practice, and on normal food intake, even high-dose dogs appear to keep from progressive potassium loss after the initial depletion. At the end of 16 months (Fig. 5), K^+ retention in the recovery period (3 days) was approximately equal to intake for that period, 72 meq. This indicates a previous depletion of about 15%, similar to that suffered the first few days of drug.

In the rat, long-term high-dose experiments did not indicate any physical sign of potassium depletion, and plasma K^+ at the end of 6 months was normal (Table IIb). We recognize the importance of muscle potassium in such experiments, but such determinations were not done.

Study of the individual experiments reveals in many cases that urinary output of K^+ was not always associated with HCO_3^- excretion, as was sodium. Figure 5, for example, shows close relation between Na^+ and HCO_3^-, but on a number of occasions (cf. day 86) there was striking loss of K^+ which appears matched by chloride. This differs from the data of Mills and Stanbury (26); they found that the rhythm of K^+ excretion in normal untreated man is inverse to that of H^+, thus more closely following HCO_3^- than Cl^-.

These variations suggest that activity of Diamox should be considered primarily not in terms of the fixed cations, but of cyclic excretion of bicarbonate and acid. It appears that the primary event in activity of Diamox is excretion of bicarbonate, and the proportions of accompanying cations depends on species, intake, and an unknown number of physiological and (in man) pathological variables. One such important variable is indicated in the experiment of Fig. 15. This dog was given 14 meq. of KCl subcutaneously each day of

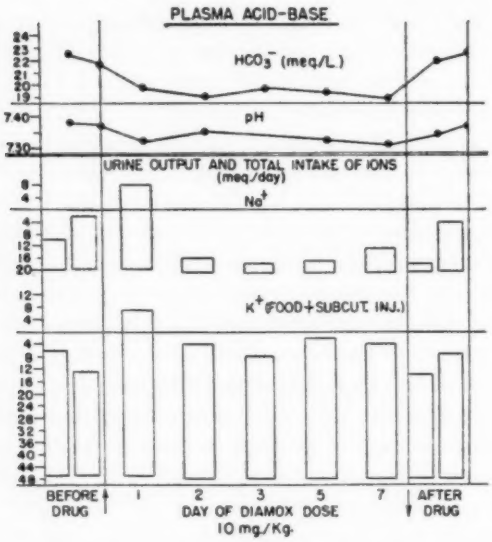

FIG. 15. Sodium and potassium balance on 10 mg./kg./day, plus K^+ supplement. Dog M 85. Wt. 10.3 kg. Fourteen meq. KCl was given subcutaneously at 9 A.M. each day, including 2 days before and after Diamox. Diamox given 9 A.M., fed 4 P.M. See Fig. 13 for explanation of balance data.

Note strikingly positive sodium balance when Diamox + excess K^+ is given.

the experiment, including the days before and after administration of Diamox. Thus, including the dietary K^+, the daily intake was 48 meq., compared to 20 meq. of sodium. When Diamox was given, there was no sodium diuresis, even in the first 6 hours. Except for the fact that all of the excreted bicarbonate was matched by potassium, the effects of Diamox were entirely comparable to the usual experiments. As seen in Figure 15, the ultimate result of this change in balance was that the dog retained sodium while on Diamox.[8] A similar situation is seen in severe cardiacs (24) and cirrhotics (28), who may tend to retain sodium, and respond to Diamox by loss of K^+ and HCO_3^-. Normal patients on a very low salt diet may show the same phenomenon (25).

3. Chloride

Traditionally, one expects regular reciprocity between chloride and bicarbonate in plasma and urine. Such a pattern was not always clear in this study.

[8] Evans *et al.* (Clin. Sc., 1954, **13**: 305.) have produced depletion of K^+ in healthy men by combination of a special diet and a sodium cation exchange resin. In this situation Diamox caused typical bicarbonate diuresis but accompanied by *greater* excretion of Na^+ and *less* of K^+ than occurs in the normal subject.

On the low-dose schedule of Figures 1 and 3 there is surprisingly little variation from normal in urinary chloride. The excess acid and ammonia excreted at night on such schedules cannot readily be paired with any one anion. Part is chloride and part is phosphate; sulfate and organic acid have not been measured. Fig. 14 shows daily Cl^- excretion on another low-dose schedule. Initially, no change from normal was seen; some decrease was seen on subsequent days, but this did not change when drug was withdrawn.

Table V gives an account of all of the ions that we have measured, on a dosage schedule (10 mg./kg./12 hours) that shows initial effect, partial compensation through acid excretion, moderate acidosis, and recovery on drug withdrawal. The role of chloride is not clear from this tabulation; on drug days some downward trend in urinary chloride is evident, but again there was no change on withdrawal. In all data on this subject it is apparent that the order of magnitude of variation (in meq./day) in urinary chloride is considerably greater than that for bicarbonate on both control days and drug days after day 1. Interpretation of changes in chloride ion is correspondingly more difficult.

On schedules (Figs. 5 and 6) that cause prolonged metabolic acidosis there is associated hyperchloremia; this elevation of 5–10 meq./l. corresponds to the lowering of plasma bicarbonate, just as in classical metabolic acidosis. Urinary chloride is so variable that no directional change is perceptible except during periods of sudden metabolic shifts. On schedules where dosage is such that acidosis occurs rapidly, there is evident at the onset some retention of chloride with slight hyperchloremia (Figs. 11 and 12). Similarly, on withdrawal from intensive treatment with Diamox, chloride excretion is fairly marked. It has already been pointed out that failure of renal response to Diamox in acidosis (data of Fig. 12) cannot be assigned to hyperchloremia, for due to vomiting on days 3–7, marked hypochloremia ensued without affecting the primary acidosis or relieving the renal refractoriness to Diamox.

In summary, it appears that shifts in chloride ion are not clearly elicited by low doses of Diamox in the dog. Chloride excretion does not play a decisive or unique role in the recovery process between doses, although it contributes considerably to the total urinary anions. On higher doses, with greater shifts in body base, a reciprocal role of chloride to bicarbonate may emerge, although even here the relationship is not entirely quantitative. It appears as if the high variability of normal chloride excretion obscures the precise relations between this ion and HCO_3^- in response to Diamox in the normal dog.

4. Phosphate

Excretion of this ion is documented in Figure 14 and Table V. It is evidently not affected by Diamox. Similar findings have been reported by others (2, 15).

TABLE V

Urinary Anions and Cations at Various Stages of Diamox Activity. Oral Doses, 10 mg./kg./12 hrs. for 6 days.

Day of Schedule	Hours after Dose	Plasma			Urine									Cations minus Anions
		pH	HCO_3^- meq./l.	Cl^- meq./l.	Flow ml./min.	pH	μeq./min. HCO_3^-	Cl^-	$(PO_4^{\uparrow})$	TA	NH_4^+	Na^+	K^+	
No. Drug A	6				.38	7.2	23	21	20	0	8	53	45	42
	12				.29	5.7	<1	46	34	15	32	18	41	26
	24	7.38	20		.15	5.8	<1	23	20	8	15	17	12	9
meq./24 hrs.							9	42	34	11	26	38	41	31
B	6				.27	6.9	12	27	14	.3	6	42	38	33
	12				.29	5.8	<1	33	34	13	8	24	33	11
	24	7.37	18	106	.14	6.3	2	12	14	4	4	11	14	5
meq./24 hrs.							5	30	29	8	8	31	35	18
C	6				.26	7.2	12	12	—	0	10	29	40	
	12				.20	5.8	<1	34	—	12	24	5	33	
	24	7.38	21	113	.12	6.2	2	14	—	3	9	8	19	
meq./24 hrs.							6	28	(31)	7	19	18	41	(20)
Drug 1	6				.63	7.9	113	11	27	0	1	89	92	31
	12				.25	5.7	<1	23	29	13	26	1	35	23
	24	7.23	13	109	.13	6.4	4	7	10	5	7	8	16	15
meq./24 hrs.							44	17	27	8	18	39	57	34
2	6				.30	7.1	17	6	29	0	7	17	56	28
	12				.26	6.0	<1	7	36	13	20	2	28	20
	24	7.26	15	120	.06	6.6	2	2	6	.5	5	3	6	4.5
meq./24 hrs.							6	6	27	5	13	9	34	22
6	6				.25	7.2	22	8	20	0	8	24	51	33
	12				.19	5.7	<1	14	27	12	41	1	7	20
	24	7.25	16	114	.15	6.2	1	16	13	4	17	3	12	7
meq./24 hrs.							8	20	27	7	30	13	29	24
Recovery 1	6				.17	5.8	<1	18	11	7	33	.7	2	13.7
	12				.21	5.6	<1	23	29	14	46	.4	8	16.4
	24	7.38	20	116	.13	5.6	<1	12	14	7	20	.2	6	7.2
meq./24 hrs.							~1	23	25	12	43	.6	8	15.6

TABLE V.—*Continued*

Day of Schedule	Hours after Dose	Plasma			Urine,									Cations minus Anions
		pH	HCO_3^- meq./l.	Cl^- meq./l.	Flow ml./min.	pH	μeq./min. HCO_3^-	Cl^-	$(PO_4^=)$	TA	NH_4^+	Na^+	K^+	
2	6				.23	6.1	<1	21	12	5	18	16	21	27
	12				.18	5.4	<1	27	23	12	23	5	15	5
	24	7.42	18	114	.09	6.4	2	5	7	2	5	9	9	11
meq./24 hrs.							1	21	18	7	18	14	19	19
3	6				.16	6.5	2	13	7	2	8	7	26	21
	12				.18	5.7	<1	11	29	11	24	7	8	10
	24	7.41	21	115	.08	6.5	2	2	7	2	7	1	7	6
meq./24 hrs.							1	10	18	6	17	6	18	18

0 time is 9 A.M. each day. Animal fed at 9 A.M. Dosed at 9 A.M. and 9 P.M. Water *ad lib.* Daily intake 18–24 meq. Na^+; 27–36 meq. K^+. Dog P 20. 12.4 kg.

* (PO_4) values are mM × 1.8, which is its base equivalence at pH 7.4. Figure in parenthesis is an average daily phosphate value for the previous days.

Plasma phosphate and calcium concentrations in long-term acidotic dogs are normal. Data and discussion are given in Ref. 1.

5. Matching of Cations and Anions

Table V indicates that the measured urinary cations minus anions are not systematically altered in various stages of administration of Diamox. Excess of cations over anions amounting to 16–34 meq./day (out of a total cation output of about 100 meq./day) is probably accountable to absence of values for two important anions, sulfate and organic acid. These two anions, in man, account for about 25% of total anion (5), which is the same percentage of the whole as the difference between measured cations and anions observed here.

The relative constancy of measured cations minus anions suggests that no new ion is formed in significant amounts. Urinary electroneutrality appears to be preserved in the various stages of Diamox activity by variation in concentration of the ions enumerated in the foregoing pages.[2]

6. Water

Data on urine flow is included in certain of the tables and figures. Variations in flow appear secondary to effects on electrolytes but do not invariably follow such changes. During the drug phase of most experiments flow is approximately twice normal. This increase may be eliminated by antidiuretic hormone; in recent experiments we have substantially *reduced* flow during the drug phase

of Diamox in dogs by intravenous injection of 5 milliunits of pitressin. Electrolyte output was not affected. It seems clear that Diamox has no primary effect on renal excretion of water.

C. Other Factors Bearing on the Renal Effects of Diamox

1. Time of Drug Administration with Respect to Feeding and Urinary Excretion Cycle

In the course of these experiments it became clear that after the initial dose of Diamox, the effects of subsequent doses spaced 24 hours apart or less were strikingly influenced by the time of day they were given and by their relation to feeding. It has already been observed [Section A.2(b)] that the response to single low doses given every 24 hours was larger and more regular when the dogs were fed at the time of dosage than if they were fasted on test days. Also, when Diamox was given every 12 hours starting in the morning the dog ***never*** responded when dosed at night without food (Fig. 8).

Figure 16 is an attempt to systematize these observations by describing the course of bicarbonate excretion in both fed and fasting dogs given 10 mg./kg. of Diamox in the morning and at night. There are four combinations to be explored in an initial drug administration, as indicated by the letters A–D for the four schedules in Frame 1 of Figure 16. These data show that the dog responds well to Diamox *initially* at either time, with or without food; there is somewhat better response in the morning with food than in the evening with fasting, just as control excretion of bicarbonate at the former time is always greater than at the latter. Diamox seems then to exaggerate the normal rhythm of bicarbonate excretion. Diurnal excretory cycles of other ions may be noted from Table V.

Frames II–IV of Figure 16 follow these schedules for several days. The final frame (V) shows how bicarbonate excretion over a 63-day period was affected by time of administration of Diamox and feeding. (This is the experiment of Fig. 8.) The data of Frames II–V may be summarized by saying that under these conditions of moderate base depletion, the ***most*** favorable circumstance for activity of Diamox is morning dosage at the time of feeding, and the ***least*** favorable is evening dosage without feeding. These differences are impressive. The other two combinations (A.M. dosage when fasting, and P.M. dosage when fed) are intermediate between these extremes. These results follow the pattern suggested (Frame I) by differences in control excretion of bicarbonate, and the small differences in response to the first dose among these four schedules.

We may conclude that the renal effect of Diamox, particularly in the face of a moderate or slight acidosis, is markedly influenced ***both*** by the feeding schedule with respect to dosage and by the time of day the drug is given.

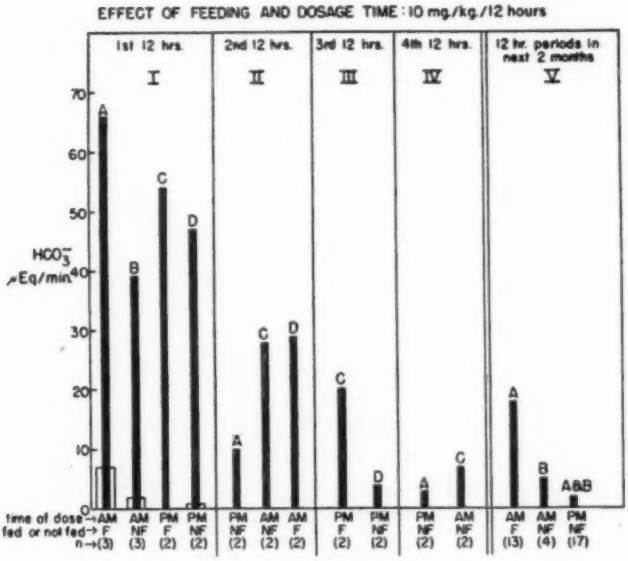

FIG. 16. Relation of feeding and time of dosage to renal effect of Diamox.
Key: A.M. = 9 A.M.; P.M. = 9 P.M.

F = fed at time of Diamox dose

NF = not fed at time of Diamox dose

⌐ = HCO_3^- output in untreated control dogs, 4–12 determinations in a total of 7 dogs

(n) = number of experiments on same or different dogs; averages are plotted.

Each letter represents a dosage schedule of Diamox of 10 mg./kg./12 hours, but starting under the 4 different situations of time and feeding depicted in Frame I. Successive frames indicate responses as dosage is continued through consecutive 12-hour periods. The dogs were fed every 24 hours, so Frame III has the same time and food conditions as Frame I, and Frame IV as Frame II. As noted elsewhere in this paper, by the second day, base depletion is fairly fixed, so that Frames III–V indicate effects in the acidotic animals, and I in the normal, with II, intermediate. Dosage B differs in that it is not a continuous schedule, but represents occasions (in V) when the dog refused to eat at 9 A.M., but appeared entirely well. Long-term effects in the acidotic animal are indicated in Frame V for one animal (that of Fig. 8) during 2 months of treatment on 10 mg./kg./12 hours. The effects of schedules A and B on P.M. excretion in Frame V are combined, since morning feeding, or lack of it, did not influence night response.

2. *Effect of Sex on Response to Diamox*

Table IIa shows that male and female rats give a similar quantitative response to Diamox, as judged by sodium and potassium excretion. However, since all of the above data for dogs are from the female, it was thought advisable to give the drug to a male dog. Following 10 mg./kg. of Diamox orally

TABLE VI

Diuretic Effect of Oral Diamox in Untreated Dog with Low Plasma HCO_3^-. 5 mg./kg. at 0 Time

Date Hours after Dose	Plasma						Urine				
	pH	pCO_2 mm Hg	meq./l			K^+	Flow ml./min.	pH	μeq./min.		
			HCO_3^-	Cl^-	Na^+				HCO_3^-	Na^+	K^+
Oct. 8, '53											
−1 to 0	7.28	27	12	105	142	3.1	.08	7.6	4	2	18
0 to 3	7.24	29	12	102	147	2.9	.37	8.1	105	43	94
Oct. 14, '53											
−1 to 0	7.33	32	16	107	—	3.4	.12	7.2	4	1	23
0 to 3	7.21	31	12	107	—	3.0	.31	8.1	87	25	100

M79, 14.8 kg. Blood collected at end of sample period. Dog not fed.

administered, 6-hour excretion of ions in microequivalents/min. were: HCO_3^-, 68; Na^+, 39; K^+, 64. Reference to the preceding data (for example, Figure 4, the same dose in a female) shows that this experiment in the male results in the same urinary pattern as noted throughout for the female.

3. Effect of Diamox on Glomerular Filtration Rate

Berliner *et al.* (2) observed a drop in inulin clearance following Diamox given intravenously. We have usually (for example, Fig. 3c in Ref. 1) seen a small drop in creatinine clearance after Diamox given intravenously. Following Diamox orally, however, no drop is observed. Figure 12 shows creatinine clearances on successive days before, during, and in recovery from high-dose treatment with Diamox. In the face of radical changes in urinary composition, creatinine clearance was not greatly changed. There was the suggestion here (and in the duplicate experiment) that with the progression of the severe acidosis, the clearance was somewhat increased.

The two dogs referred to in the above paragraph (experiment of Fig. 12) were subsequently used for chronic toxicity studies at 33 mg./kg./day. Their history is recorded in Table IX of Ref. 1. During the course of this long-term treatment and resulting moderate acidosis, creatinine clearance was determined on four occasions. In all cases it was within the normal range for these dogs, again with a suggestion of upward trend (Section D.3 of Ref. 1). Thus, it appears that long-term treatment with Diamox does not greatly alter glomerular filtration rate.

4. Effect of Diamox in "Respiratory Alkalosis" with Low Plasma HCO_3^-

It has been shown that Diamox does not give a renal effect in the face of base depletion, and that plasma HCO_3^- concentration is not the controlling factor (Fig. 11).

Further evidence that plasma bicarbonate concentration is not a primary determinant of the activity of Diamox is available from Table VI. Quite by chance it was found that the dog of this experiment, on at least two occasions, had low plasma bicarbonate and normal pH without prior treatment. Correlated with these data is the behavior pattern of the dog; when handled and caged it showed marked hyperpnea. Presumably this is the pattern of hyperventilation or "respiratory" alkalosis. Despite plasma bicarbonate concentration as low as 12 meq./l., it showed typical renal response to Diamox. In a 15-kg. dog such a plasma concentration (8 meq./l. below normal) could reflect loss of bicarbonate from as little as 6 to as much as 80 meq., depending on whether the plasma sample is considered to represent plasma alone, all extracellular fluid, or total body water.

In recent acute experiments (14, 14a) in which there was fairly constant filtration of bicarbonate, we have observed that initially Diamox causes excretion of 20% of filtered HCO_3^-, but this decreases to 2% with increasing loss of total body bicarbonate.

Thus, diverse lines of evidence point to net loss of total body HCO_3^- as the primary factor in renal refractoriness to Diamox.

SUMMARY AND DISCUSSION

The results of renal carbonic anhydrase inhibition as illustrated with Diamox are an initial *diuresis* of Na^+, K^+, HCO_3^- and water, and an *acidosis*, the extent and type of which depends on size and frequency of dose.

The character of the diuresis has been described in Section A.1. It is emphasized that this diuresis is almost entirely independent of the dose of Diamox above 5–10 mg./kg. This suggests that inhibition of renal carbonic anhydrase is maximal at this dose range.

Documentation of details of the acidosis and its effect on the diuretic action of Diamox form a large part of the present communication. It has been shown that the acidosis is both metabolic and respiratory in nature. The metabolic component appears to predominate, and it is this component that appears responsible for the altered response to Diamox. In the course of this work it was found essential to define quantitatively the extent of the acidosis on various dosage schedules of Diamox. This could not be done reliably in terms of plasma pH, pCO_2, or HCO_3^-. It has been noted that low plasma bicarbonate can occur with almost any degree of total bicarbonate deficit, while the same change in pH can result from either a metabolic or respiratory acidosis. On the other hand, the *total base economy* of the body, as defined in the Introduction, provided a useful and reproducible means of measuring departures from normal acid-base balance.

The relation between renal diuretic response and metabolic acidosis is summarized in Table VII. This summary presents the salient points of all

TABLE VII

Summary of Base Depletion and Bicarbonate Excretion on Various Dosage Schedules Following Diamox

Dose	Duration of Drug Adm., days	Total No. of Dogs or Individual Dog Nos. and Text Reference	n§	Feeding Schedule on Test Day†	HCO_3^- Excretion 4 or 6 Hrs. after Drug μeq./min.		Base Depletion during Treatment meq/kg‖.
					Av	*Range*	
0		4 dogs	10	Fed	11	(1–23)	—
0		9 dogs	17	NF	6	(1–27)	—
Single Doses							
5–100 mg/kg		4 dogs	5	Fed	111	(68–137)	—
5–100 mg/kg		9 dogs	16	NF	84	(51–165)	—
Continued Dosage* (mg/kg/time)							
10 mg/48 hrs.	11	P47 p. 293	3	NF	50	(47–51)	0
5 mg/24 hrs.	20	M84 Fig. 3	4	Fed	47	(41–56)	0.5
	17	P47 p. 293	12	NF	25	(8–45)	
	31	M73 p. 293					
10 mg/24 hrs.	29	P20 Fig. 4	6	Fed	60	(27–81)	1.5
	7	A42 p. 293	4	NF	17	(11–21)	
10 mg/12 hrs.	63	M14 Fig. 8	14	Fed	33	(13–58)	2.8
	6	P20 Table V					
	63	M14 Fig. 8	4	NF	8	(1–13)	
100 mg/24 hrs.	470	M73 p. 294	5	Fed	32	(9–55)	3.9
	471	P27 p. 296					
	470	M73 Figs. 5–7	15	NF	9	(1–23)	
	471	P27 p. 296					

* Data on bicarbonate excretion in this section do not include day 1. Determinations were made on random days throughout drug administration.

§ n = number of tests of HCO_3^- excretion.

† Fed = dog fed at time of dosage. NF = not fed.

‖ Calculated from excess acidification on recovery, see procedure 2 of Introduction. Values represent averages of data from similar dosage schedules.

experiments discussed above, in terms of these two major effects. Comparison of Column 7, base depletion, with Column 6, bicarbonate excretion, clearly indicates the conclusion of these studies: *The greater the acidosis, the less the diuretic effect.* On low doses (5–10 mg./kg./day) there is stimulation of renal acidification, so that the total excretion of acid each day is above normal (cf. Fig. 9, line A). It is this process that appears to maintain acid-base balance near normal and preserves the diuretic effect. On high single (100 mg./kg.) or multiple small (10 mg./kg./12 hrs.) daily doses there is an extremely variable pattern of daily acidification which does not generally exceed the normal value; acid-base balance is never permanently restored, and the continued administration of Diamox results in sustained acidosis. In this

situation (3–4 meq./kg. base depletion) the diuretic response is either sharply diminished or at best erratic.

There are rather striking differences in the responses of dogs, depending on whether they are fed or fasted on the test day. In the latter situation, they are more sensitive to acidosis and will not give a typical diuretic response in the presence of even small imbalance. Repetition of diuresis is achieved in the fasted animal with 10 mg./kg. spaced every 48 hours. In the fed animal, there is good response to Diamox at the same dose every 24 hours in the presence of a mild (about 1 meq./kg.) base depletion.

There are no radical changes in plasma or urinary electrolytes as dosage is prolonged from several weeks to 16 months. This supports the idea that in chronic acidotic dogs the initial depletion of base is simply not restored, and it is this depletion that accounts for the final deficit. Further corroboration of this thesis may be found by comparing the initial losses (Table IV) with the final depletion in base economy (Table VII).

The dynamic situations in the various dosage schedules considered, from 5 mg./kg./day to 33 mg./kg./8 hours, are strikingly different. Yet the fundamental reaction of the animal is the same in all; the result is the prevention of continuing loss of base, which would rapidly be fatal. On low daily doses, extra base output is effectively countered by extra base conservation during recovery periods. On high or multiple daily doses, treatment renders the animal refractory to the diuretic effect itself; no further changes occur.

Initial challenge with Diamox given around the clock (Fig. 11) produces depletion of at most 8 meq./kg. (see Table IV), or approximately 100 meq. of buffer base in one day. This appears to be the limit of net HCO_3^- loss that can be elicited by Diamox under normal conditions. However, the usual rate of restoration of HCO_3^-, or extra acidification, is about 25 meq./day, indicating that it takes four times as long to regain acid-base balance as it does to lose it. The same time relation appears in single-dose studies, such as that of Fig. 1. When drug is maximally active for 4 to 6 hours, as it is following 5 mg./kg., about 18 to 20 hours are required to restore acid-base balance to normal. This relationship may be useful in formulating dosage schedules in the alternative situations where continuous inhibition of carbonic anhydrase (with acidosis and no repetitive diuresis) is desired, or where discontinuous inhibition (with no acidosis, and with repetitive diuresis) is the goal.

Certain other constant values have emerged from these experiments. Base depletion over 3 meq./kg. leads to markedly diminished response to Diamox, and depletion over 5 meq./kg. is associated with virtual abolition of response. We have found that the same amounts of added acid produce similar effects (14). Following Diamox, it appears that the dog will deviate permanently to the extent of 5–8 meq. base loss/kg., but no further. Such animals (Figs. 5, 8, 11, 12) are "deadlocked" in acidosis, but show no abnormality other than

the chemical (1). The "deadlock" results from the fact that there is no further diuretic response, and at the same time Diamox prevents recovery, i.e. extra acidification of urine. A reasonable inquiry now is: Why does the animal, which is refractory to the diuretic effect of Diamox, not take the final step and overcome the acidosis by extra acidification while still on Diamox? The probable answer to this emerges from current experiments (14). In *extreme* acidosis, following NH_4Cl, the typical repressive action of Diamox on extra acidification *can* be overcome. We have observed NH_4^+ plus acid output five times normal in the presence of Diamox, but only when the acid load following NH_4Cl was as great as 12 meq./kg. At that level of acidosis, Diamox has no effect on the kidney. In the 3–8 meq./kg. range, either of base depletion following Diamox or of administered NH_4Cl, the inhibitor exerts some physiological effect, i.e. prevention of extra acidification and subsequent recovery of acid-base balance. We are tempted to believe that the dog is physiologically indifferent to base depletion in this range, since the animal does not overcome it and can live with it comfortably. Under the challenge of considerably higher acidosis, however, the dog excretes extra acid in the presence of Diamox, suggesting a reserve mechanism independent of carbonic anhydrase (14, 14a).

It may be emphasized that these studies are introductory, and attempt only to establish a baseline for certain renal effects that seem to typify responses to carbonic anhydrase inhibitors in the *normal* experimental animal. It is hoped that future work will provide answers to the many related questions still outstanding in this field.

CONCLUSION

1. The renal effects of a single oral dose of Diamox in the normal dog are described. These occurred in sequence as follows: (A) There was diuresis of HCO_3^-, Na^+, K^+ and H_2O when plasma concentrations of Diamox remained above about 2 γ/ml. (B) When Diamox concentration in plasma declined below this level, there was retention of these ions, with extra urinary excretion of hydrogen and of ammonium ions until balance was restored.

2. The magnitudes of (A) per unit time were largely independent of dose (and plasma concentration) above 5 mg./kg. in both rat and dog. This suggests that maximal inhibition of carbonic anhydrase was achieved. Increasing the amount of a single dose maintained the diuretic phase (A) as long as plasma concentration of drug remained above 2 γ/ml.

3. When small (5–10 mg./kg.) daily doses were given to normal dogs the diuretic phase (A) lasted about 6 hours, and the recovery period (B) about 18 hours. In this situation acid-base balance was preserved, since the animal has had enough time in (B) to excrete acid equivalent to the base lost in (A). Each day when Diamox was given, therefore, the dog's electrolyte status was roughly that of day 1, and it responded to Diamox as on that day.

4. When higher single doses or multiple doses were given each day, the time available for recovery was shortened; as drug-free time lessened, the acidosis became greater. Response to Diamox under these conditions was diminished or, in severe acidosis, entirely lost. There was neither recovery nor further base depletion; after as long as 16 months of continued treatment, the dog had about the same acidosis that it had on day 2. The base depletion in high-dose regimens studied has been of the order of 5 meq./kg.

5. The acidosis, chiefly metabolic in type, includes a respiratory component and thus may be considered a "mixed" acidosis. It is suggested that this respiratory element is due to slight inhibition of blood carbonic anhydrase.

6. The evidence from these and other studies suggests the view that total body acid-base balance determines response to Diamox. Excess of acid over base diminishes the diuretic action of Diamox regardless of whether acid was gained or base lost. This depression of the action of Diamox is roughly proportional to excess acid over base in the range 1–8 meq./kg. Plasma HCO_3^- concentration *per se* did not correlate well with either base depletion or diuretic activity of Diamox.

7. Chloride ion exhibits a sluggish reciprocity to bicarbonate in these experiments. Diamox does not appear to affect phosphate excretion in the dog.

8. The evidence suggests that the primary effect of Diamox is on bicarbonate excretion. The K^+/Na^+ ratio during bicarbonate diuresis varies according to species, base intake, and pathological state. Where either cation is present in excess of normal, it is generally the predominant one in urine.

9. Variations in feeding schedule and time of dosage suggest that Diamox has maximal activity when given in the morning with food, and minimum activity at night without food.

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SOME HEMODYNAMIC EFFECTS OF THE HUFNAGEL OPERATION FOR AORTIC REGURGITATION

Studies in Models and a Patient

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Hufnagel (1) has described a surgical procedure which is intended to ameliorate the circulatory derangement of aortic regurgitation. A ball-valve is placed in the descending thoracic aorta just beyond the mouth of the left subclavian artery. This mechanism blocks diastolic regurgitation from that portion of the arterial tree which lies beyond it but presumably offers little resistance to forward flow through it during ventricular systole.

In models and in a patient, observations were made bearing on the following questions:

(1) What is the effect of the operation on the mean diastolic pressure in the segment of the arterial tree proximal to the ball-valve? (This segment supplies, of course, the coronary and cerebral systems.)

(2) What is the effect of the operation on the volume of regurgitation at the aortic valve and therefore on the work-load of the left ventricle?

(3) What is the effect of the procedure on the diastolic murmur of aortic regurgitation?

(4) What are the quantitative inter-relationships between these three parameters—diastolic pressure, volume of regurgitation, and size of murmur—before and after operation?

(5) To what extent can the change in the murmur and in diastolic pressure be used clinically to evaluate the degree of reduction in volume of regurgitation and, in general, the effects of operation?

OBSERVATIONS ON MODELS

Methods. A sketch of the type of model which was constructed for use in these studies is presented in Figure 1. Rubber tubing with an internal diameter of ½ inch and a wall thickness of $\frac{3}{32}$ inch was used to represent the arterial system. One end (analogous to the site of capillary run-off) was blocked. At the other end (analogous to the aortic valve area) the barrel of a syringe was placed in the system. The orifice of the syringe tip, measuring about $\frac{1}{32}$ inch in internal diameter and ½ inch in total length, represented the orifice through which leak occurs at the aortic valve in aortic regurgitation. The narrow orifice was selected to permit detailed study of the pressure-volume-murmur-time inter-relationships by spreading out the time scale. (In the second set of experiments, the effective orifice was further reduced by the addition to the syringe tip of a 20-gauge needle 1½ inches long.) With a clamp applied at A the water-filled system was inflated to a "systolic" pressure of 300 mm. Hg. (Only

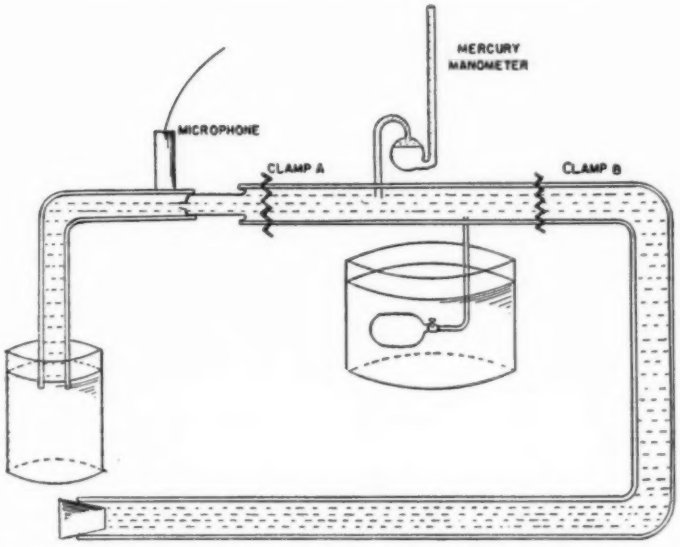

FIG. 1. Diagrammatic representation of the model used in these studies. See text for detailed description.

"diastole," not "systole," was studied in these experiments.) Then the clamp was released at A and "regurgitation" allowed to occur through the syringe tip orifice. At 5 or 10 second intervals, observations were made on the volume of "regurgitation" and on the "diastolic" pressure in the proximal segment. The murmur produced during "regurgitation" was also recorded on magnetic tape by means of a condenser-type microphone (Altec 21B150), which was applied with fluid-coupling to the surface of the tube just beyond the syringe tip. The intensity of the murmur at separate points in time was read directly from the meter of the recording amplifier and expressed in arbitrary units. In this model, *the Hufnagel operation was simulated by clamping the tube at B, thereby preventing "diastolic regurgitation" from the distal segment.*

Two main experiments were performed. In the first, the model was arranged precisely as indicated in Figure 1 and two sets of observations were made (see Table I):

(1) With both segments (total volume = 500 cc.) open for "regurgitation" through the syringe in "diastole" ("pre-operative" state).

(2) With the distal segment (volume = 385 cc.) blocked for "regurgitation" during "diastole" ("post-operative" state).

In the second experiment observations of "diastolic" pressure and volume of "regurgitation" were made, but the murmur was not recorded. Furthermore, a 20-gauge needle 1½ inches long was added to reduce the effective size of the leaking orifice; in this way it was possible to make more detailed observations on the time course of "regurgitation" since it was spread out even more than in the first experiment. The following four sets of observations were made (see Table II):

(1) With proximal and distal segment analogs of equal volume (each 41 cc.).

(a) With the entire system open for leak in "diastole" ("pre-operative").

(b) With the distal segment analog closed for leak in "diastole" ("post-operative").

TABLE I

First Set of Experiments

Time	"Pre-operative" (Vol. = 500 cc.)		"Post-operative" (Vol. = 115 cc.)	
	Volume of Regugitation	Pressure	Volume of Regurgitation	Pressure
0 sec.	0 cc.	300 mm.	0 cc.	300 mm.
5 sec.	27	190	23	58
	28	192	20	62
	26	187	22.5	64
10 sec.	60	120	35	27
	55	120	33	27
	60	120	35	26
	58	122		
	58	124		
15 sec.	80	73		
	83	74		
	80	68		
20 sec.	100	42		
	95	42		
	105	48		

(2) With the volume of the distal segment analog (61.5 cc.) approximately three times that of the proximal segment analog (20.5 cc.), a set-up which will be referred to below as the "25–75 system". (This relationship is probably more comparable to that in the human subject (6).)

(a) With the entire system open for leak in "diastole" ("pre-operative").

(b) With the distal segment analog closed for leak in "diastole" ("post-operative").

All the volumes given above were volumes of fluid in the rubber tubes used as analogs of the arterial system and were measured at atmospheric ("zero") pressure.

In a third set of experiments a 12-foot length of rubber tubing with an internal diameter of ¾ inch and a wall thickness of ⅛ inch was used to study volume of "regurgitation" in complete detail; "regurgitation" to a "diastolic" pressure of zero was permitted to occur in order to study the proportionality of the several total volumes of "regurgitation."

Results. The validity of the model is suggested by the realistic appearance of the murmur as displayed by spectral phonocardiography (Fig. 2). (This method of sound portrayal (2) displays frequency spectrum on the ordinate and time on the abscissa with intensity represented by degree of blackness in any given portion of the record.) It is not clear what significance can be attached to the presence of two harmonics. Musical or "sea-gull" aortic diastolic murmurs demonstrate this feature characteristically (2).

Table I presents the data derived from the first set of experiments. The same data are presented in graphic form in Figures 3–5. "After operation" the "diastolic" pressure in the proximal segment fell lower in a given period of time than "before operation" (Fig. 3). Furthermore, the volume of regurgitated fluid was decreased "after operation" (Fig. 4). Although the volume of

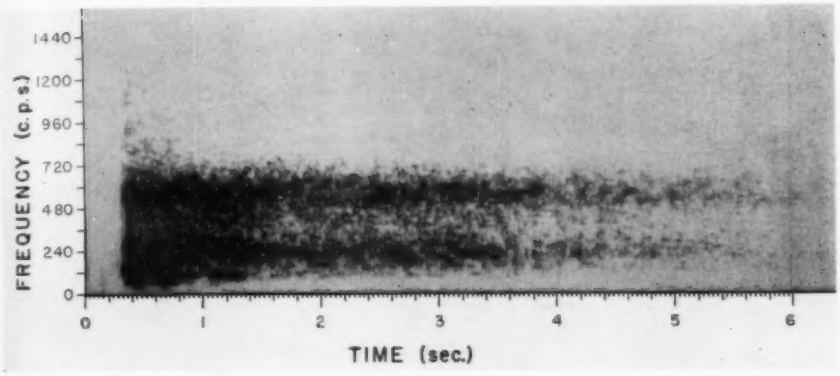

FIG. 2. Spectral phonocardiogram of the murmur produced at the syringe tip in the model diagrammed in Figure 1.

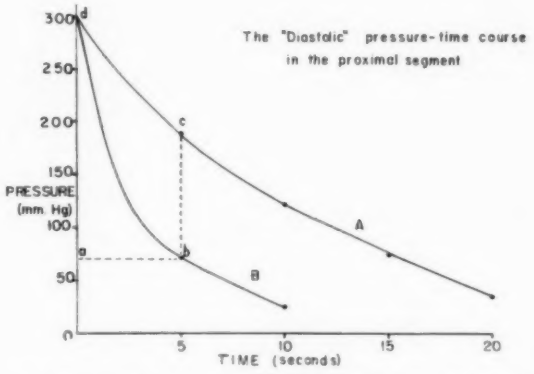

FIG. 3. "Diastolic" pressure before (A) and after (B) "operation" in the first set of experiments.

total "regurgitation" was reduced by "operation", the volume of "regurgitation" in 5 seconds from the proximal segment was increased from 7.5 cc. (6.5% of the static volume of the proximal segment) to 34 cc. (30% of the static volume). The "operation" reduced the size[1] of the murmur in the first five seconds of "diastole" to approximately 42% of the "pre-operative" value. The last numerical expression was arrived at by a comparison of the area of *abd* with that of *abcd* (Fig. 5). The ratio of the areas under the corresponding curves in the pressure-time plot is 46% (Fig. 3). These figures suggest a close inter-

[1] Size of murmur is the area under an intensity-time plot; that is, it takes both intensity and duration into account. The unit of expression for murmur size can be decibel-seconds, millivolt-seconds, or dynes-per-square-centimeter per second.

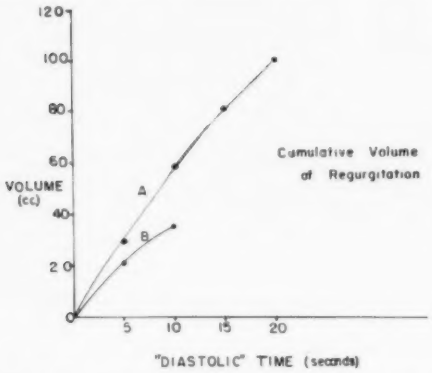

FIG. 4. The volume of "regurgitation" before (A) and after (B) "operation" in the first set of experiments.

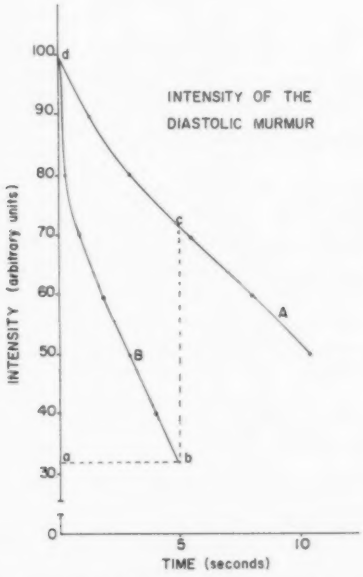

FIG. 5. Size of "diastolic" murmur before (A) and after (B) "operation" in the first set of experiments.

dependence of "diastolic" pressure and the size of the murmur. No such agreement between the ratio of murmurs and the ratio of volumes of "regurgitation" was attained, the value for the latter ratio being 78% at the five-second point (Fig. 4).

TABLE II

Second Set of Experiments

Time Seconds	"Pre-operative"		"Post-operative"	
	Regurgitation Volume (cc)	"Diastolic" Pressure (mm. Hg) In Upper Segment	Regurgitation Volume (cc)	"Diastolic" Pressure (mm. Hg) In Upper Segment
Upper Segment, 140 cc.; Lower Segment, 140 cc. ("50-50" system)				
0	—	300	—	300
10	10.8	246	10.5	218
	10.8	242	10.6	214
20	9.6	196	8.2	152
	9.5	200	8.2	150
30	8.5	154	6.2	98
	8.2	158	6.3	100
40	7.0	116	4.4	64
	6.8	122	4.3	62
50	5.4	88	2.8	40
	5.4	90	3.0	40
60	4.4	66		
	4.4	68		
Upper Segment, 70 cc.; Lower Segment, 210 cc. ("25-75" system)				
0	—	300	—	300
10	11.	245	10.3	192
	11.	252	10.2	190
20	9.6	196	7.3	110
	9.3	207	7.0	113
30	8.7	162	4.4	62
	8.2	154	4.6	64
40	6.8	124	2.6	45
	6.5	118	2.6	46
50	5.8	92		
	5.5	86		
60	4.3	70		
	4.2	64		

Table II presents the data derived from the second set of experiments. The same data are presented in graphic form in Figures 6 and 7. "After operation" the "diastolic" pressure in the proximal segment fell lower in a given period of time than "before operation". On the other hand, the volume of "regurgitated" fluid was decreased "after operation". Both of these effects of "operation" varied inversely with the ratio of the volume of the proximal segment to the volume of the distal segment. Although the volume of total "regurgitation" was reduced by "operation", the volume of "regurgitation" from the proximal segment was *increased* by "operation". For instance, in the "25–75 system" approximately 9.0 cc. (one-fourth of 36 cc.) regurgitated from the proximal segment in 40 seconds "before operation" whereas "after operation" 25 cc.

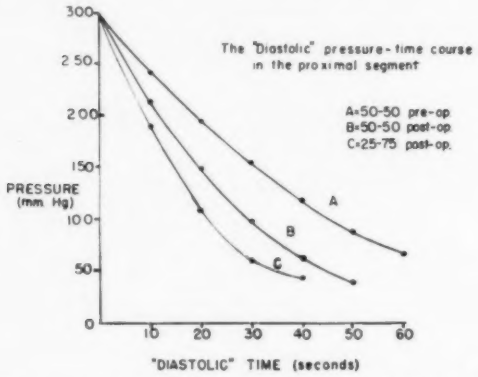

FIG. 6. "Diastolic" pressure before (A) and after (B and C) "operation" with two different ratios of proximal segment to distal segment.

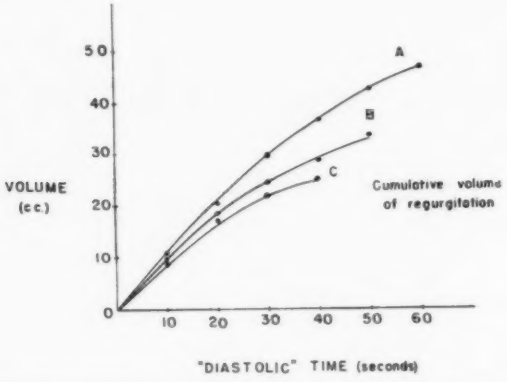

FIG. 7. The volume of "regurgitation" before (A) and after (B and C) "operation", with two different ratios of proximal segment to distal segment, in the second set of experiments.

or almost three times as much "regurgitated" in the same period of time. These "regurgitant" volumes are 12% and 33%, respectively, of the static volume of the proximal segment. The volume regurgitated in the first portion of "diastole" changes relatively little "after operation". The longer "diastole" was, the greater was the relative reduction in the volume of "regurgitation" after "operation". (This last feature is best demonstrated by the third set of experiments in which "regurgitation" was permitted to proceed to completion, i.e., to a diastolic pressure of "zero". These results are shown in Figure 8. It will be noted that the total volumes of "regurgitation" do bear a direct proportional relationship to the volume of the system from which "regurgitation" is occurring. In Figure 9 the cumulative volumes of "regurgita-

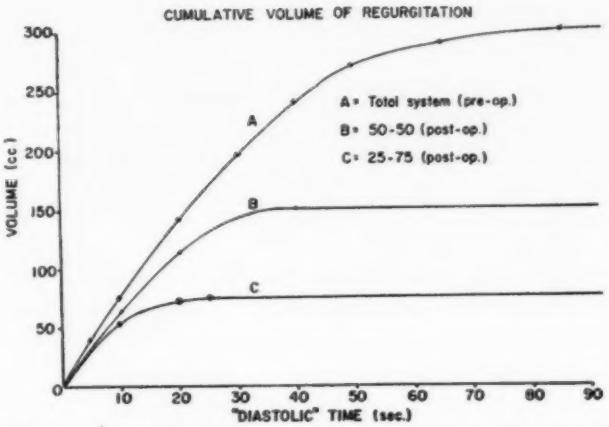

FIG. 8. Volume of "regurgitation" in the third set of experiments in which "regurgitation" was permitted to proceed to completion, i.e., to a "diastolic" pressure of zero.

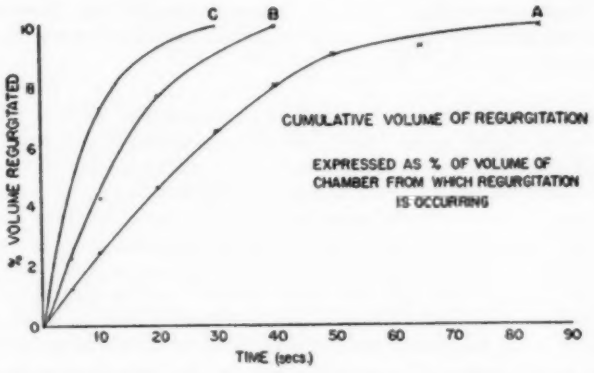

FIG. 9. The volumes of "regurgitation" in the third set of experiments expressed as percentages of the resting volume of the chamber from which "regurgitation" was occurring.

tion" are expressed as percentages of the total "resting" volume of the chamber from which "regurgitation" occurred.) Figure 10 presents the data on "diastolic" pressure from the second set of experiments recalculated as mean pressure for "diastolic" periods of various durations.

OBSERVATIONS IN A PATIENT

Methods. B. G., a 35-year old colored male, had the physical signs of "pure" aortic regurgitation, presumably on the basis of rheumatism. The development of incapacitating dyspnea prompted consideration of the Hufnagel procedure. Calibrated records of heart sounds on magnetic tape were made before and after operation under identical conditions.

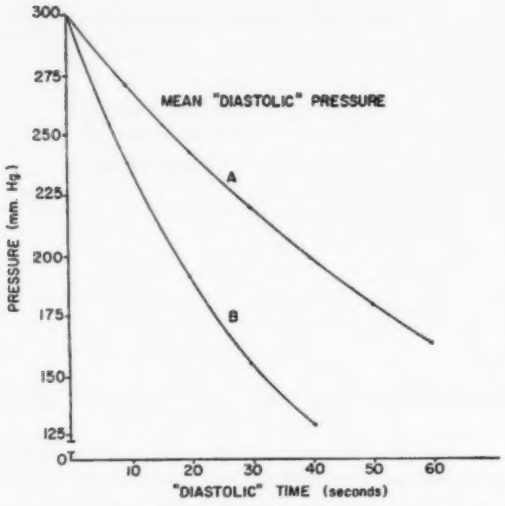

FIG. 10. Graphic presentation of data on "diastolic" pressure (from the second set of experiments) recalculated as mean pressure for "diastolic" periods of various durations. A = before "operation". B = after "operation".

At operation[2] similar calibrated sound recordings were made from the base of the aorta and from the surface of the left ventricle before and after the placing of a Hufnagel ball-valve in the descending aorta just beyond the mouth of the left subclavian artery. These recordings were made on magnetic tape using the microphone described above. Before the insertion of the valve, a calibrated pressure pulse recording was made from the ascending aorta and afterward from both the ascending aorta and the descending aorta distal to the ball-valve. Changes in stroke volume were estimated by the roentgenkymographic method. The difference (in square centimeters) between the systolic and diastolic areas of the heart as determined in roentgenkymograms in the PA and LAO projections was taken as the Stroke Volume Index and was compared before and after operation (3). The areas were measured by planimetry.

Results. The pressure tracings made in the operating room are shown in Figure 11. As seen in Table III, diastolic pressure and mean pressure (particularly mean diastolic pressure), measured proximal to the artificial valve, *fell* after operation. The diastolic hypertension distal to the valve after operation suggests marked vasoconstriction in the tributaries of the lower aortic segment. This was almost certainly the result in part of the period of occlusion of the aorta.

Before operation, the Stroke Volume Index was 25.8, as measured in the PA projection and 24.9 as measured in the LAO projection. Fifteen days after operation, the Stroke Volume Index was 22.6 as measured in the PA projec-

[2] Operation was performed by Dr. Henry T. Bahnson.

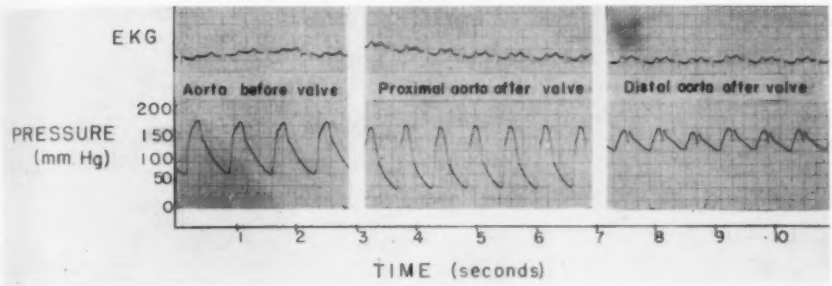

FIG. 11. Aortic pressure before and after Hufnagel procedure in patient B.G. Recorded directly at thoracotomy.

TABLE III

Pressure Measurements in Operating Room in Patient B. G.

	Pressure	Mean Pressure*	Mean Diastolic Pressure*	Mean Systolic Pressure*	Pressure at Incisura	Duration of Systole	Duration of Diastole	Intervals	
								R to Upstroke	R to Incisura
	mm. Hg	*mm. Hg*	*mm. Hg*	*mm. Hg*	*mm. Hg*	*sec.*	*sec.*	*sec.*	*sec.*
Aorta before operation	180/75	117	90	151	135	0.30	0.42	.07	.36
Proximal aorta after operation	175/35	96	46	141	90	0.285	0.30	.085	.37
Distal aorta after operation	175/135	157	—	—	148	0.28	0.31	.15	.36

* Determined by planimetry.

tion. Three months after operation the value was 12, a value which appears to represent marked decrease in volume of regurgitation.

Of incidental interest is the spectral phonocardiogram (Fig. 12) of the loud wooden sounds produced by the artificial valve *in situ* and recorded from the immediate vicinity of the valve at operation. The sounds have high frequency components. Distinct splitting of both the "first" and the "second" sound is present. As will be discussed later, this splitting is of interest in connection with splitting of the normal heart sounds.

Figure 13 presents the relationship of the artificial valve sounds to the normal heart sounds and to the elements of the electrocardiogram. The valve opens about 0.07 sec. after the arterial valves open (as indicated) approximately by S1. The difference between the closing times of the arterial and artificial valves is not great. These findings are corroborated by the study of the relationship between the EKG and the pulse pressure tracings proximal and distal to the artificial valve (see Fig. 10 and Table III).

Figure 14 presents a comparison of the aortic diastolic murmur before and after operation. These are intensity-calibrated spectral phonocardiograms (2),

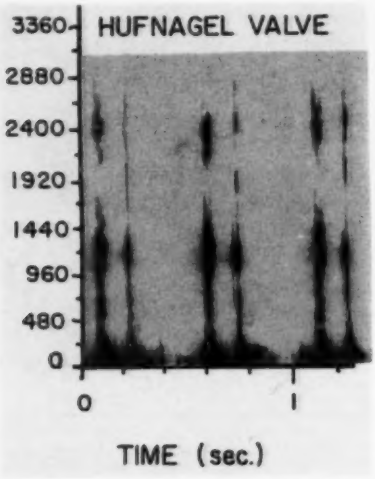

FIG. 12. Spectral phonocardiogram of the sounds produced by the artificial valve. Recorded from vicinity of valve at thoracotomy.

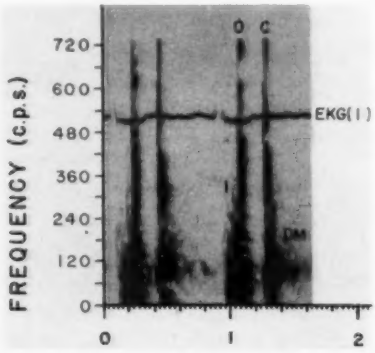

FIG. 13. Here is shown the relationship of the opening ("o") and closing ("c") sounds of the artificial valve to the normal valve sounds and to the elements of the electrocardiogram.

which were recorded and analyzed in an identical fashion and are therefore comparable. The systolic murmur changed little, a good control. Marked reduction in the diastolic murmur is demonstrated. Not only is its intensity (blackness) much less but its frequency range is more limited.

Clinically it is difficult to be sure of over-all improvement in the patient and at least one new symptom has developed since operation. Dizziness has been present but is difficult to evaluate. Angina persists. Dyspnea is not pronounced, but the patient restricts his activity sharply and follows closely the various

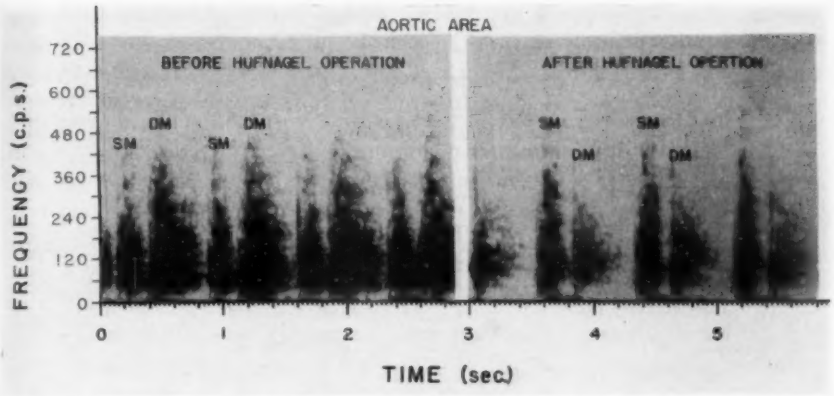

FIG. 14. Graphic display of the decrease in size of aortic diastolic murmur after operation as compared with the pre-operative finding. See text for comments.

details of a cardiac regimen. He voluntarily points out that he no longer has the profuse sweating he characteristically had before operation. The new symptom is blurring of vision in the left eye with ophthalmological evidence of early optic atrophy and early cataract formation on the left. Since cataract and optic atrophy are manifestations of reduced blood supply to the head, as in the aortic arch syndromes (7), it is being entertained that the eye findings are related to operation. No other explanation is now evident. Since operation the intra-ocular pressure of 15 mm. Hg has been sufficient to cause collapse of the retinal arteries in diastole. In future cases these observations should be compared before and after operation. (We are indebted to Drs. R. E. Hoover and F. B. Walsh for the ophthalmologic observations in this patient.)

As measured with the standard sphygmomanometer, blood pressure in the arm before operation averaged 180–200 mm. Hg systolic with change in tone of the Korotkoff sounds at about 40 mm. and persistence of sounds to the zero pressure level. Since operation systolic pressure in the arms has been in the range of 230–240 mm. Hg and no change in tone has been present in the Korotkoff sounds on reducing the pressure to zero. Blood pressure in the legs, measured with a wide 12 cm. cuff., averaged 150/80 mm. Hg at a time when systolic pressure in the arms was much lower than usual, 160/0.

Pulse rate and therefore length of diastole has an important bearing on what occurs following operation. (With short diastolic periods one can expect, from the model experiments, relatively less reduction in regurgitation by operation.) The patient's average pulse rate in a basal state in the hospital before operation was approximately 75 per minute. After operation the average of some thirty observations by the patient himself under basal conditions,

such as those on first waking, in the morning, was approximately 82 per minute.

DISCUSSION

It is clear that the diastolic pressure head for perfusion of the cerebral and coronary circuits is reduced after operation. One can anticipate a decrease in cerebral and coronary blood flow and possibly some symptoms referable thereto. Syncope is uncommon in aortic regurgitation in contradistinction to aortic stenosis and probably no great increase in this manifestation can be expected after this operation. Giddiness in the upright position has apparently been a frequent occurrence in the post-operative (5) period and has prompted later ambulation in these patients than would ordinarily be practiced. This phenomenon, although difficult to interpret when it occurs after a major surgical procedure such as this, is given adequate explanation by the experiments reported here.

The two physiological effects of operation can be expected to have opposing effects as far as angina is concerned. The fall in mean diastolic pressure will certainly reduce the supply of oxygen to the myocardium. On the other hand, the reduction in volume of regurgitation will reduce its work and therefore the demand for oxygen. Whether angina will develop or be aggravated by this operation will depend on which way the balance of supply and demand is tipped.

Two factors might favor the passage of an increased proportion of the stroke volume to the tributaries of the aorta proximal to the artificial valve. These factors are (1) the fact that aortic pressure must attain a level of at least 80 mm. of mercury before the ball-valve opens whereas a negligible level of diastolic pressure is present in the proximal aorta; and (2) the resistance to flow created by the ball-valve itself. It may be that in practice the increase in systolic flow to the proximal segment of the circulation may offset in part the effects of the operation on diastolic flow in this same segment.

Rough estimation of the relative volumes of the arterial tree proximal and distal to the point where the valve is implanted leads to the conclusion that approximately 75% of the total arterial volume is beyond this point. As far as length of aorta proximal and distal to this point, Remington (6) found that in 48 aortas that 22% of the total length (on an average) was made up of proximal segment. However, as is clearly demonstrated by the observations in both the models and the patient, it by no means follows that by the operation "approximately 75 per cent of the reflux is controlled" (1). It is true that if diastole is very long, the reduction in volume of regurgitation approaches this value (see Fig. 8). Due to the differences in its wall, the proximal segment of aorta is a more effective energy reservoir than the distal segment and for that reason also a further deviation in the form of greater regurgitation from the proximal segment is to be expected.

These experiments indicate a reason, even more compelling than the technical one, why the ball-valve cannot be placed in the ascending aorta. In that situation the regurgitant leak would occur from such a small chamber that the coronary arteries would, in effect, derive their blood directly from the ventricle at the low, almost negligible, level of diastolic pressure.

It will be very pertinent to measure cerebral and coronary blood flow before and after operation in these patients. The comparison of A-V oxygen differences, before and after operation, for the cerebral circuit and for the coronary circuit will be valuable. Otis (8) points out that good samples from the superior vena cava and inferior vena cava usually show close correlation as to oxygen content. This might not be the case after implantation of the artificial valve if blood flow to the area drained by the superior vena cava suffers or is not improved commensurate with that in the lower part of the body.

Confirmation of predictions as to the effect of operation on diastolic pressure in the proximal segment has been provided by the observation of Rose and collaborators (4) in a series of patients operated on by Hufnagel. In nine patients studied before and after operation, diastolic pressure in the brachial artery was lower after operation in every case and, with the exception of a single case, systolic pressure was higher. The experiments described here would, however, lead one to an interpretation quite different from theirs: "The concept of more complete left ventricular emptying following operation also provides an explanation for the higher systolic and lower diastolic pressures in the regurgitant brachial artery seen after as compared to before operation."

As we have shown by means of models, regurgitation from the proximal segment is exaggerated after operation. The reason that the systolic hypertension of aortic regurgitation is even more pronounced after operation is the same as the reason that systolic hypertension occurs at all in aortic regurgitation. Furthermore, the reason that the pressure peak in the proximal segment is even sharper (with more striking "systolic collapse") after operation (4) is the same as the reason that these phenomena occur at all in aortic regurgitation. The pattern of left ventricular ejection is altered in aortic regurgitation. The low diastolic pressure results in rapid ejection of most of the stroke volume early in systole. As a result, aortic pressure rises to an early high peak and falls rapidly later in systole. The systolic hypertension may be due in part to the increase in stroke volume, but is due even more to a drastic change in the shape of the systolic portion of the arterial pulse pressure curve. It is unlikely that mean systolic pressure is much altered.

Most of the peripheral signs of aortic regurgitation—carotid dance, DeMusset's bobbing of the head, capillary pulse, pistol-shot sounds, etc.—are dependent more on the derangement of the systolic phase than that of the diastolic (9). They are the result of the change in the shape of the systolic

ejection curve and of the resulting aortic pressure curve. These signs disappear, of course, in the lower part of the body following operation. They may be exaggerated and almost certainly will not be diminished in the upper portion of the body.

The splitting of the sounds which are produced by the impact of the valve ball against its seating in both the open and the occluding position is perhaps to be expected. In closing, the ball might be expected to impinge on one side of its circular seat and then on the rest of the seat. Similarly an asynchronous impact of the ball on the corners which hold the valve open in cardiac systole is not surprising. It seems conceivable that minute splitting of the normal second heart sound may be the result of minor asynchronism in the coaptation of the separate cusps of the arterial valves. It is to be noted (10) that spectral phonocardiograms display splitting better than classical oscillographic phonocardiograms. The spreading-out of the frequency spectrum results in display of distinct components at some if not all points. In oscillograms a merging of the components is likely to occur unless the splitting is relatively pronounced.

CONCLUSIONS

(1) The implantation of a ball-valve in the descending thoracic aorta in cases of aortic regurgitation results in *decrease* in the volume of aortic regurgitation, a *decrease* in mean diastolic pressure in the segment of the arterial tree proximal to the ball-valve and a *decrease* in the size of the diastolic murmur.

(2) The size of the diastolic murmur correlates well with mean diastolic pressure in the proximal segment and poorly with the volume of regurgitation.

(3) Both the beneficial effect on volume of regurgitation and the adverse one on mean diastolic pressure are greater the smaller the segment proximal to the ball-valve; i.e., the nearer the artificial valve is to the base of the aorta.

(4) The exaggeration (by the operation) of the depression of mean diastolic pressure in the proximal arterial segment (which includes the cerebral and coronary systems) may limit the over-all benefits of the operation.

(5) Splitting of the sounds produced by the artificial valve is demonstrated by spectral phonocardiography. Asynchronous impingement of the valve on its seatings is suggested as the mechanism of this minute splitting.

(6) The opening and closing sounds of the artificial valve *in situ* occurs between the normal heart sounds. Systole is abbreviated distal to the artificial valve.

(7) In a patient in whom the Hufnagel procedure was performed, diastolic pressure was decreased in the proximal segment and systolic pressure increased. Ocular manifestations of possibly ischemic origin have apparently developed since operation. The size of the aortic diastolic murmur and the volume of regurgitation (as indicated by roentgenkymography) have decreased since operation.

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THE CLINICAL FEATURES ASSOCIATED WITH KIMMELSTIEL-WILSON LESIONS

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A study has been made of certain clinical characteristics of patients with diabetes mellitus and of the correlation of these findings with the presence of Kimmelstiel-Wilson lesions in the kidney at autopsy. Such study has allowed a definition of the clinical picture associated with proven Kimmelstiel-Wilson lesions, as contrasted with the clinical features of diabetic patients who at autopsy had renal lesions of other types or who had no renal lesions. Certain differences in the metabolic patterns of these groups have been reported (1). It is our present purpose to record in similar comparative fashion other clinical features of the three groups with particular reference to the symptom complex arising in association with the Kimmelstiel-Wilson lesion.

METHODS

The methods used in the study of these patients have been described (1), and only a brief recapitulation will be given here. The records of all patients with diabetes mellitus who were autopsied at the Johns Hopkins Hospital in the years 1938 to 1951 were reviewed. The clinical features of these patients were recorded and, without knowledge of the findings, the pathologist, Dr. Stanton Eversole, examined histologic sections of the kidneys. This permitted an unbiased comparison to be made of the clinical features presented by patients (1) with diabetes and proven Kimmelstiel-Wilson lesions, (2) with diabetes and renal disease of other types, and (3) with diabetes but without significant renal pathologic change.

The pathologist's sole criterion for the diagnosis of the Kimmelstiel-Wilson lesion was that described by Kimmelstiel and Porter (2) as a hyaline nodular mass in the glomerulus. The terms Kimmelstiel-Wilson lesion and glomerular nodule are used interchangeably in this report, and the process is called glomerular nodulation. For reasons discussed in a previous paper (1) the term intercapillary glomerulosclerosis has not been used. An attempt has been made to estimate quantitatively the extent of the Kimmelstiel-Wilson lesions, and of arteriolar and arterial changes in the kidneys. It is recognized that such quantification cannot be precise. Arteriolosclerotic nephritis was diagnosed by the pathologist when he found hyaline arterioles in the kidney with involvement of glomerular arterioles causing narrowing and varying degrees of hyalinization of the glomeruli.

The criterion used for the diagnosis of diabetic retinitis was the presence of a moderate number or of many capillary aneurysms seen on ophthalmoscopic examination during life or in a few cases in a flat preparation of the retina following autopsy. These aneurysms often appear as multiple, deep, punctate hemmorrhages during life but when examined histologically are shown to be focal dilatations or aneurysms of the capillaries in the bipolar cell layer of the retina (3). Exudates and hemorrhages, because they may arise from

other causes, were not considered diagnostic of retinitis unless accompanied by characteristic descriptions of sharply circumscribed red spots. We are aware that in this type of reconstructive study errors in the interpretation of the retinal examination must have occurred.

Obesity was diagnosed if the examining physician so stated. Diabetes mellitus was diagnosed if the patient had glycosuria and fasting blood sugar greater than 130 mgm. per cent. Onset of diabetes was dated from time of unequivocal symptoms or from the first recorded chemical proof of its presence. It is apparent that such estimates, especially with reference to patients with mild diabetes, may not be representative of the total duration of the metabolic defect.

In the construction of Table III it was obviously unwise to decide whether the peripheral edema was primarily renal or cardiac in origin. Arbitrarily "heart failure" was diagnosed if concomitant with the peripheral edema, the patient was noted to have symptoms or signs of pulmonary congestion, or of raised peripheral venous pressure.

RESULTS

These are shown in Tables I to V. In Table I there is a single significant difference among the groups as regards age of onset, namely, that only in the group with renal diseases of other types has there been a considerable number of individuals with the onset of diabetes after the age of sixty years. The age of onset does not otherwise differentiate the three groups, since more than half of each group had the onset of diabetes between the ages of thirty-one and

TABLE I

Age of Onset and Duration of Diabetes in All Patients Coming to Autopsy 1938–51 Who Had Diabetes Mellitus and Glomerular Nodules

	With Glomerular Hyaline Nodules	With Other Renal Lesions	With No Renal Lesion
Number of Patients	45	48	85
Number of Patients Classified According to Age at Onset of Diabetes.			
<15 years	2	3	4
16–30 years	5	0	12
31–45 years	14	11	20
46–60 years	16	15	37
>60 years	4	16	11
Unknown	2	3	1
Number of Patients Classified According to Duration of Diabetes			
<3 years	1	13	38
3–5 years	4	11	16
6–10 years	15	11	15
11–20 years	17	9	10
>20 years	4	2	5
Unknown	2	2	1

sixty years. It is noted that in the entire study there are too few cases of juvenile diabetes to provide a valid sample.

In Table I there also appears a classification of the duration of diabetes in the three groups. Only a few patients developed glomerular nodules within six years of the apparent onset of the diabetes. In two patients diabetes was not demonstrated. These patients had no clinical history suggesting diabetes, and no sugar was detected on examination of the urine. Neither of these patients had studies made of fasting blood sugar or of their glucose tolerance. One patient had retinal aneurysms and the pancreas of the other patient showed hyalinization of islet tissue.

In the two groups without Kimmelstiel-Wilson lesions, approximately half of the patients had five years or less of known diabetes at the time of death. It is possible that some or all of these could have developed Kimmelstiel-Wilson lesions at a later date. Nevertheless, there were fifty-two patients with diabetes of duration six years or more, and twenty-six patients with diabetes for eleven years or more, who did not develop the glomerular lesions.

In Table II is shown the comparison of clinical findings in the three groups. No single characteristic sets off definitively the patients with glomerular nodules from those patients without nodules.

In Table III is shown the occurrence of "heart failure" (see Methods) in the patients of the three groups at the time that edema was present. It is noted that in the patients without Kimmelstiel-Wilson nodules, the edema almost always was accompanied by cardiac failure. In contrast in those patients shown to have Kimmelstiel-Wilson nodules edema was often present

TABLE II

Comparative Study of Characteristics of Patients with and without Glomerular Nodules

	With Glom. Nodules				Without Glom. Nodules	
	Mild	Moderate	Marked	Total	With renal disease	Without renal disease
Number of Patients	13	26	6	45	48	85
Number of Patients Showing:						
Diabetic Retinitis	9	20	6	35*	6†	6‡
Obesity	10	19	5	34	26	38
Proteinuria > 2+	7	16	6	29	18	20
Edema > 2+	6	14	6	26	7	3
Diastolic B.P. > 100 mm. Hg	9	14	5	28	20	19
Retinitis plus Edema	7	13	6	26	1	2
Azotemia at Death NPN > 70 mgm%	5	12	6	23	21	11
Triad: Edema, Hypert. and Alb.	5	9	5	19	4	1

Lack of Information * in 5 Patients, † in 6 Patients, ‡ in 9 Patients.

TABLE III

Occurrence of Heart Failure in Patients with Edema

Group	Total Patients	Edema	Other Evidence of Heart Failure At Time of Edema
Kimmelstiel-Wilson	45	30	14
Renal Lesions of other Types	48	7	6
No Renal Lesion	85	3	3

TABLE IV

Comparison of Clinical Characteristics of All Patients with Kimmelstiel-Wilson Nodules and of Control Patients with Diabetes of More than 5 Years Duration

	With Nodules	With Renal Disease	Without Renal Disease
Number of Patients	45	24	31
Retinitis	35	6	2
Proteinuria	29	12	4
Hypertension	28	14	6
Edema	26	7	2
Retinitis plus Edema	26	1	0
Azotemia	23	11	3
Triad: Proteinuria, Hypertension, Edema	19	3	0

without evidence of congestion in the pulmonary or peripheral venous circulations.

Retinal aneurysms and peripheral edema in combination appeared in the same patient in twenty-six of the forty-six patients with Kimmelstiel-Wilson nodules, but in only two per cent of the controls. The symptom complex of hypertension, albuminuria and edema was present in less than half of the patients with glomerular nodules. When present, because of the infrequency of these combinations in the control groups, retinitis and edema, or edema, albuminuria, and hypertension constituted good positive evidence of the coexistence of glomerular nodules. Since patients who have died with less than five years of diabetes may not have had time to develop Kimmelstiel-Wilson nodules, these data are presented again in Table IV. It can be noted that the control patients with more than five years of diabetes show a much higher incidence of the individual abnormalities of hypertension, proteinuria, retinitis, and edema. It is still true, however, that the combination of retinitis and edema or of edema, hypertension and albuminuria, is uncommonly found in the absence of glomerular nodules.

From the classification of the Kimmelstiel-Wilson process according to the severity of the lesions in histological study, it is noted that in the six patients in whom this process was extensive, six exhibited retinitis, edema, severe

TABLE V

Clinical Features of Patients Showing Kimmelstiel-Wilson Lesions Associated with Minimal Vascular Nephritis

K. W. Severity	Age at Death	Duration of Diabetes in Years	Retinal Aneurysms	Azotemia at Death	Edema	Proteinuria	B.P. mm. Hg.
+	58	8	No	No	No	No	165/110
+	60	15	No	Yes	No	Yes	155/75
++	60	15	Yes	No	No	No	160/80
+	37	10	No	No	No	No	125/70

proteinuria and terminal uremia. Five of the six patients had diastolic hypertension, and the sixth patient had blood pressure readings of 225 systolic, 85 diastolic. When the Kimmelstiel-Wilson process was mild or moderate in extent, the classical symptom complex developed in about one-third of the patients.

As stated under *Methods*, attempts were made to estimate the severity of arteriosclerosis and arteriolosclerosis in the kidneys of these patients and to make correlation of these data with the severity of the Kimmelstiel-Wilson process as noted in the sections. Satisfactory quantification of these vascular diseases was not always possible, but it can be stated that in the kidneys of each of the six patients with marked Kimmelstiel-Wilson lesions there was extensive arteriosclerosis and arteriolosclerosis. In the thirty-nine patients with less marked Kimmelstiel-Wilson involvement, the amounts of arteriosclerosis and arteriolosclerosis were variable. Of the fourteen patients in these two categories who developed the combination of edema, albuminuria and hypertension, all had evidence of marked or moderate arteriosclerosis and arteriolosclerosis of the kidneys. There were four patients with Kimmelstiel-Wilson involvement but with minimal disease of the arteries and arterioles. The clinical picture of these patients is shown in Table V. Although the sample is small, it is interesting to note that none of these patients had triad of edema, albuminuria and hypertension or the combination of edema and retinitis.

A study was made in the patients with Kimmelstiel-Wilson nodules of the frequency of occurrence of the complete nephrotic syndrome according to the criteria used by Bloom and Seegal (4). These were serum albumin, less than 3.0 Gram per cent; cholesterol, greater than 300 mgm. per cent, edema, and proteinuria. Although many of the patients had proteinuria and edema, only two patients had co-existent high cholesterol and low serum protein. It must be recognized however that the data were incomplete in some instances. A more systematic survey of this problem is needed.

DISCUSSION

The recognition of the glomerular nodule as the criterion for the diagnosis of the renal disease due to diabetes mellitus, has permitted in recent years a

better definition of the clinical features of this entity (2). The clinical features associated with the renal lesion as presented in this paper do not differ in their larger outlines from those of recent studies (2), (5–10) which have utilized the same criterion. The discussion will be limited to a few points which have not received much attention.

The six patients with maximal involvement of the glomeruli by the Kimmelstiel-Wilson process had extensive co-existent arteriolosclerotic and arteriosclerotic nephritis. In these patients the generally recognized syndrome of albuminuria, edema and hypertension* was present. Lesser degrees of glomerular nodulation had occurred in four patients with minimal arteriosclerosis and arteriolosclerosis of the kidney, and in these patients the clinical combination of albuminuria, edema and hypertension did not appear.

Although the hypertension and albuminuria are natural concomitants of arteriolosclerotic nephritis, it cannot be concluded that the characteristic massive edema of patients with extensive glomerular nodulation is the result solely of arteriolosclerotic nephritis. As Bloom and Seegal (4) have shown, marked edema is not encountered in arteriolosclerotic nephritis in the absence of cardiac failure. The limited conclusion that can be taken from our study is that the occurrence of the marked edema in patients with the Kimmelstiel-Wilson lesion and without cardiac failure, is associated with the combination of glomerular nodulation and arteriolosclerotic nephritis. A gross analogy may be made with the one other renal disease characterized by marked edema, hypertension and proteinuria, namely, chronic glomerulo-nephritis. In the latter disease there also occurs combined involvement of the glomeruli and the arterioles, although of course the glomerular lesions are morphologically and presumably etiologically distinct from the process of glomerular nodulation.

As has been noted by others, the Kimmelstiel-Wilson process and diabetic retinitis are frequently found in the same patient (2, 3, 6, 13). The extensive evidence (12, 14, 16) that an early lesion in retinitis is the capillary aneurysm raises the possibility that the beginning process in the glomerulus may be of similar nature. Hückel, (12) Allen, (15) Friedenwald, (3) Ashton, (14) and Hall (16) mention such glomerular capillary dilatations in patients with glomerular nodules. In this series we have demonstrated such dilatations (Plate I) within the glomerular capillaries. The significance of these glomerular dilatations in terms of a single entity of capillary disease of the retina and kidney cannot be assessed at this time. In this group of patients co-existence of retinitis and glomerular nodulation was common. However, the indirect method necessarily utilized in this study for the diagnosis of retinitis prevents the application of this apparent correlation too literally. This deficiency also limits the usefulness of retinitis alone as a help in the diagnosis of the presence of glomerular nodulation. Some ophthalmologists (17) point out that single

* One patient had a blood pressure of 225 systolic, 85 diastolic.

retinal aneurysms may be found in many diabetic patients if the search is made frequently and carefully. Moreover, only a fraction of the aneurysms seen microscopically are large enough to be revealed by the ophthalmoscope (18). The transformation of these aneurysms into the often devastating disease of diabetic retinitis, and their relationship to the dilatations and nodular destruction of the glomeruli are matters for further study.

SUMMARY AND CONCLUSION

1. A report has been made of the clinical characteristics of forty-five patients shown to have Kimmelstiel-Wilson nodules at autopsy. These clinical features have been contrasted with those of one hundred and thirty-three diabetic patients who were proven at autopsy not to have Kimmelstiel-Wilson nodules.

2. The data on duration of diabetes and the age of the patient at onset of diabetes bring out no remarkable differences between the Kimmelstiel-Wilson group and the controls, except that those patients who die after less than five years of diabetes, rarely exhibit nodules. It is also noted that the development of nodules is not a simple function of length of duration of diabetes, since twenty-nine patients had diabetes for eleven years or longer but had no glomerular nodules.

3. Diabetic retinitis was present in thirty-five of the forty-five patients with Kimmelstiel-Wilson lesions and in only twelve of the one hundred and thirty-three control patients. The combination of retinitis and peripheral edema was a reliable diagnostic point in favor of the presence of glomerular nodules since only two per cent of the control diabetic patients had these signs concomitantly.

4. The classical triad of hypertension, albuminuria and edema occurred in less than half of the patients with Kimmelstiel-Wilson nodules but when present was strong evidence in favor of the diagnosis of Kimmelstiel-Wilson nodulation.

5. The co-existence of arteriosclerosis and arteriolosclerosis of the kidneys with Kimmelstiel-Wilson nodulation occurs frequently. However, glomerular nodulation can occur in the absence of significant arteriosclerosis and arteriolosclerosis. The four patients with such pure glomerular nodules did not manifest the combination of hypertension, albuminuria and edema.

6. Six patients with the most marked nodulation of the glomeruli had during life severe edema, hypertension, proteinuria and uremia. In addition, each had extensive arteriosclerotic and arteriolosclerotic nephritis. These latter diseases in pure form are not generally associated with marked edema in the absence of cardiac failure. This study points out that patients with Kimmelstiel-Wilson nodulation often develop edema not explicable on the basis of cardiac failure.

7. Dilatations of the glomerular capillaries similar in appearance to the retainal aneurysms have been noted in eleven patients with Kimmelstiel-Wilson nodules.

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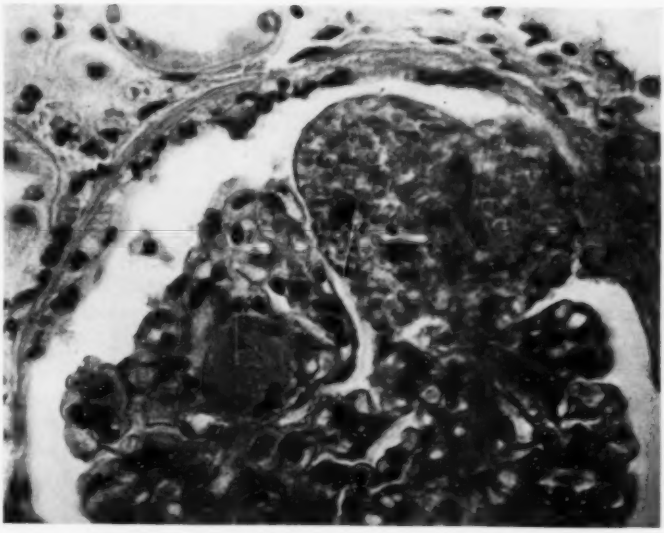
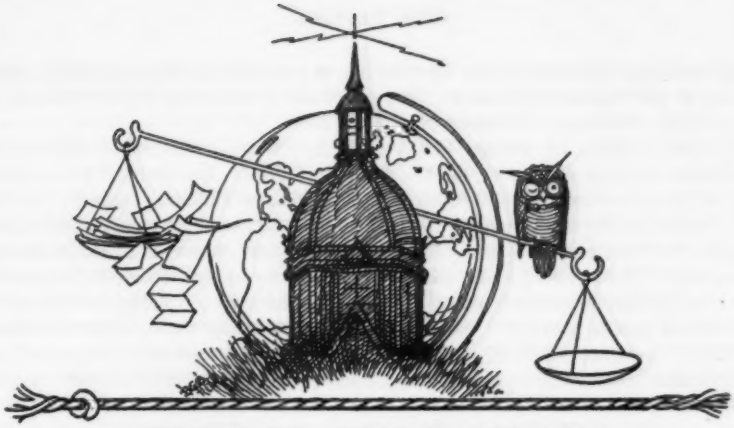
PLATE I

Photomicrograph (magnification 100×) of a glomerulus showing the hyalinized Kimmelstiel-Wilson nodule and also a blood-filled capillary aneurysm. By careful search the transition from the capillary aneurysm to the hyalinized nodule can be found in many cases.





BOOK REVIEWS

(These reviews represent the individual opinions of the reviewers and not necessarily those of members of the Editorial Board of this Bulletin)

(These reviews represent the individual opinions of the reviewers and not necessarily those of members of the Editorial Board of this Bulletin)

Geriatric Medicine, 3rd edition. Edited by EDWARD J. STIEGLITZ. 718 pp., $15.00. J. B. Lippincott Co., Philadelphia, Pa.

This book was first published in 1943, revised in 1949 and is now again revised. The editor recognizes that "no one physician is competent to be authoritative in all the many aspects of geriatric medicine"—(which is) "not a specialty in the usually accepted sense—. It is rather the application of the understanding of the changes which come with sensecence to all of the facets of medical practice."

In the first section of the book are twelve chapters concerned with the scope of geriatric medicine in general, the anatomical and physiological changes of aging, the general medical, psychiatric and surgical care of aging patients. The subsequent seven sections cover various diseases, divided according to the system involved, in the special light of the problems introduced by aging and senescence. There are forty-seven contributors in addition to the editor.

It has not been possible for the authors to cover their subjects in detail because of space limitations, but their discussions are uniformly of good quality, with sound therapeutic recommendations, and there is a bibliography after each chapter which offers further information to those interested. There is an excellent index.

The editor states that "geriatric knowledge is needed by all who see older patients, whether as general practitioners or as specialists". The former group will find this volume very helpful, and the latter group may profit considerably from the large first section which will help them give the aging individual a life with "depth and breadth as well as length".

ROBERT E. MASON

Science and Man's Behavior. The Contribution of Phylobiology. Including the complete text of The Neurosis of Man. By TRIGANT BURROW. Edited by William E. Galt. 564 pp., $6.00. Philosophical Library, New York, N. Y.

This book is edited by William E. Galt, Ph.D., Research Associate of The Lifwynn Foundation, of which the author was the Scientific Director. The author "was unable to bring the project to completion" because of untimely death. The second section, The Neurosis of Man, An Introduction to a Science of Human Behavior, should undoubtedly be read first. It "contains the essence of" the phylobiological researches of the author and his associates. The foreword states that "phylobiology is the application of the scientific method to the field of human relations". Through "social analysis" the author has studied "disorders of human behavior", which are to him "essentially social or interrelational". He evolved "a science of feeling" with special attention to "our internal motivations" and "an organismic factor that is as yet unrecognized by man". He believed that "disorder resides within the tissues of phylic man" and he concerned himself with the nature of the human organism, considering it in "the stark immediacy of the moment" as studied by "my impartial senses." This research "arose quite spontaneously from the recognition that neurosis is pandemic to man and that not only my patients, but I myself, my family and my friends were necessarily part of this phylic disorder. The motive, therefore, to my phyloanalytic undertaking was no other than to escape this discomfort common to me, to those about me and to the world at large. . . . " Instead of the normal he talks about "the social reaction-average", and then implies that all of society is presently abnormal, suffering from a *social neurosis*, with discriminating, selfish attitudes, wars and so on.

To appreciate the subject it is necessary to devote time to learn the new words the author developed and used. The glossary lists sixty-eight terms originated by the author. Only in this manner can one really understand his concept of the 'I'-persona, which has developed a faulty process of attention called *ditention*, and therefore is partitive, autopathic, ditentive, the product of affecto-symbolic, socio-cortical function. This function he identifies with a third or symbolic nervous system. The goal of phylobiology is to achieve cotention, "the organism's primary pattern of integration", which "results in direct, unprejudiced and cooperative behavior". This is expounded at length, often repetitively, in Part I, Interrelational Man, and Part II, Organismic Man.

The author is at odds with the dichotomy of "right" and "wrong" and feels that "Through a biological approach to man's behavior as a phylum, the allegorically conceived notion of the Fall of Man as a descent from virtue into sin and his filial resources to repentance and amendment will be replaced by his application of a mature scientific principle to his phyloörganismic rehabilitation. The ignorance and superstition of metanomic thinking, or of the numen, will become clearly manifest; and orthonomic thought, or the nomen, will assume supremacy over man's processes as a species or phylum". The religious concept of mankind is thus replaced by a material one.

The first section of the book is an assemblage of interesting comments and questions presented by scientists with whom the author communicated by forwarding to them advance material on the book which consisted in an outline and some of the key chapters. Biologist Ralph S. Lillie said, "You have shown skilfully and convincingly how deep-seated a disparity there now is between the biological requirements of man as an animal species and the characteristics and propensities of its individual members. During evolution there seems to have been an overdevelopment of the self-regarding as distinguished from the group-regarding characters of the species; hence a condition has arisen which you describe as a conflict between the claims of the 'I-persona' and the real interests of the human phylum; this conflict may well prove a source of disaster unless our scientific knowledge of

human behavior advances sufficiently, on both the theoretical and applied sides, to make possible effective preventive measures." On the other hand, philosopher George Boas wrote "I am not sure that I understand what you mean by 'normality' ". Psychiatrist Leland E. Hinsie commented "Evidently I cannot *feel* your thesis as you do, though I think many have been on the periphery of it". Psychologist David Katz says "If I understand you correctly, cotention is the final attitude towards which you strive. But can any human being survive with a breathing rate of about 4?" It should be noted that these men had access to only about one-third of the final product, and this third held promise of discoveries and principles "which throw new light on the sphere of human interrelations". This reviewer does not find that the promise bears fruit in the reading of the whole book.

Apparently the author experienced a change in attitude concerning the way he looked at life and found it so moving that he felt it represented a cure-all. But one is led to inquire, once the author achieved the physiologic state of cotention (and evidently several of his associates did too), what happened next? How did the group function differently? We are not told. In fairness to the author, he did state that this work represented only "a very meager beginning" and that a great deal remained to be done. Perhaps his co-workers have carried on and could answer some of the questions inevitably raised in an unbiased consideration of this work.

The appendix presents a very interesting series of physiologic observations concerning changes in respiration, ocular movements, basal metabolic rate and electroencephalogram as one passes from ditention to cotention and back.

EDWARD L. SUAREZ-MURIAS

Traumatismos Medulares. Clinica Y Tratamiento. By VICTOR SORIANO. 357 pp. Editorial Labor, S. A. Argentina. Buenos Aires, Montevideo.

This monograph, written in Spanish, is based on the extensive experience of the author in the field of spinal cord trauma resulting from twelve years of close contact with patients at the Institute of Traumatology at Montevideo.

In nineteen chapters the author reviews the clinical, pathological and etiological factors involved in the various levels of spinal cord trauma. Considerable emphasis and detail are given to the chapters on rehabilitation of the motor and urinary functions. These chapters set forth many of the principles of Donald Munro. Three chapters, 8, 9, and 10, are devoted mainly to clinical case reports which go into excessive detail.

In the chapter dealing with therapy the author takes no definite stand as regards cord decompression and merely presents previously stated principles. In the chapter pertaining to the clinical signs of cervical trauma he presents dermatome charts, many of which differ from the usual texts. In the chapter on the relief of pain and spasticity the author presents accepted methods of therapy. The very important aspects of metabolic and endocrinological abnormalities are described briefly and the author rather superficially presents the psychological aspects of cord trauma. Although the final chapter is limited to the discussion of regeneration it omits some of the current experimental work on this subject.

The bibliography is of international scope and covers quite well the current literature. The index is in chapter form which limits the subject reference value. The book is well bound and the publishing and printing are of high quality. The diagrams are clear and convey well their meaning. The monograph does not add appreciably to the knowledge of spinal cord trauma but is a valuable review and compilation of the current teachings on this subject.

GEORGE W. SMITH

Clinical Management of Behavior Disorders in Children. By HARRY BAKWIN AND RUTH MORRIS BAKWIN. 495 pp., $10.00. W. B. Saunders Co., Philadelphia, Pa.

Intended primarily for the physician with a pediatric practice—and hence best suited to his needs—this excellent text can be commended to all workers interested in the psychology and psychopathology of childhood. Its authors, already well known to the pediatrician, are thoroughly steeped in knowledge of children's behavior disorders. To this basic attribute are added good clinical judgment and felicity of style. Particularly worthy of mention are the sections on: The Newborn; Growth and Development; Emotional Deprivation; Disturbances of Eating; and Cerebral Damage and Behavior Disorder.

Throughout, the pediatrician is sensitized to those factors in the biological and environmental spheres likely to be related to given patterns of disturbance. These then provide the foundation for rational therapeusis, in so far as this is available. The practical aspects of management are stressed. An excellent feature in this regard is a well chosen list of books and self-help organizations for parents of children afflicted with mental retardation, blindness, cerebral palsy and other defects. The discussion of parental attitudes is comprehensive and sympathetic, though at times oversimplified. The reader may thus be left unprepared for the complexity and difficulty of altering certain pathological parental attitudes. But perhaps this belongs within the province of the psychiatrist. The authors do have a tendency to cite uncritically studies of limited value and often of a mutually contradictory nature. A bibliography is available for the reader who wishes to pursue the matter further, but it seems to me that an author has a responsibility to evaluate, not just to cite, lest the reader be abandoned in the misty night of eclecticism.

Apart from minor differences with the authors here and there, inevitable in the present unsettled state of child psychiatry, the one major consideration with which I would take issue is the relative naivete displayed toward cultural factors. The authors occasionally attribute differences in behavior or intelligence to sex, race and nationality, where these differences are largely explicable by cultural conditioning. It is a fact that Jewish children do tend to come out higher, and Negro children lower, on intelligence tests, as the authors state. But inadequate mention is made of the profound consequence of cultural heritage, economic circumstance and social opportunity on these very tests. The correlation between borderline mental deficiency in children and their parents depends at least as much upon environmental as upon presumptive genetic factors. The differences in behavior between boys and girls, while based on a biological sex difference, is certainly in large part attributable to the social pressure for conformity to culturally prescribed patterns. This is clearly shown by the studies on pseudohermaphrodites at this hospital by Dr. Wilkins' group. The child's psychological "masculinity" or "femininity" appears to depend upon what he is raised to be and not on his genetic, physiologic or anatomic equipment. In general, not enough weight is given to social factors as etiologic agents in children's behavior difficulties. I venture the hope that this deficit will be corrected in the next edition of what I confidently expect to be a much demanded text.

I cannot conclude without mention of the attractive format provided by the publishers and the happy choice of a frontispiece, a magnificent portrait of "Mother and Child" by Pablo Picasso.

LEON EISENBERG

The Dynamics of Virus and Rickettsial Infections. International Symposium. Edited by FRANK W. HARTMAN, FRANK L. HORSFALL, JR. AND JOHN G. KIDD. 461 pp., $7.50. The Blakiston Co., New York, N. Y.

Three blind men examined an elephant. In this case the elephant is called "Virus and Rickettsial Infections". The first examiner is a student of phages and is equipped with a

biochemical laboratory. With the rapid and great advances in techniques and with the brilliance of the group working on this problem this first examiner has been able to describe well that part of the problem. But the behavior of these parasites of bacteria includes lysogeny; perhaps more fundamental to our knowledge of parasitism than all of the work on T_2-T_4 etc. And lysogeny is given little attention in this particular symposium.

The second blind man is the plant virologist. He has been studying virus infections for a long time. It was he who first demonstrated filterability, inclusion bodies, interference phenomena, who purified viruses and has some knowledge of the structure of the agents he is studying. The essay of Commoner on Plant Viruses and Proteins demonstrates that progress is continuing.

The third examiner—supreme only in his homocentric position—is the animal virologist. He is just beginning to develop the tools with which to study his part of the elephant. This is illustrated by Hirst's careful "Studies on Double Infection with Influenza Virus", and by the occasional reference to newer methods of titering viruses and to the use of new cell-virus systems.

This symposium has been arranged, however, as if it now were time to integrate and correlate the things into one model. Would the penetration of a spear into the hide be similar to that of a piece of food into the mouth? And yet we find them all lumped together.

It is a valuable symposium in that it has published the careful analysis of Hershey, Luria, S. Cohen, among the phage men, of Kunkel, Bawden, Commoner among the plant virologists, and of Hirst, Shope, Price, Bodian, Habel, K. F. Meyer, Weller and others among the animal virologists.

It does not seem to this reviewer, however, that the symposium was successful in putting together a real picture of the dynamics of virus infections. This is, no doubt, because it is too soon.

FREDERIK B. BANG

Behandlung Innerer Krankheiten. Richtlinien und Ratschläge fur Studierende und Ärzte. By FERDINAND HOFF. 608 pp., Ganzleinen DM 55. ($13.10). Georg Thieme Verlag, Stuttgart, Germany.

Since this therapeutic guide is in its fifth edition, there is no doubt about its popularity in Germany. *Behandlung innerer Krankheiten* reads easily, and its recommendations are usually specific. The text is written with authority, but without more than casual reference to the published literature (a flaw to be found in *Current Therapy*.) Elaborate therapeutic regimens are described for disorders for which there are currently believed to be no specific remedies. It seems needless to review the various programs, but characteristic of the detail are such recommendations as the use of yogurt and curds in the treatment of cirrhosis of the liver. Except to fulfill one's curiosity about the practice of medicine in Germany, there is little to recommend this book to the American physician.

O. D. RATNOFF

The Female Sex Hormones. By LEON J. DEMERRE. 219 pp., $3.50. Vantage Press, Inc., New York, N. Y.

This is a rather unusual book to review, for although it does not lack appeal, it is difficult to know what type of reader would be attracted by a text of this sort. The author in his preface makes the very honest statement that "there are two types of books on the subject of sex hormones. The first is highly technical and is intended for the specialist or the student of endocrinology. The second type emphasizes and sensationalizes sex, often to the subordination of interest in sex hormones, frequently at the expense of scientific truth".

He then goes on to state his intent to achieve an intermediate type, appealing to the layman, but not deviating from the scientific truth, and intended "in the large therapeutic interests of the general public". It is a little difficult for this reviewer to understand how such a text will make any monumental contribution to public therapeutic interest, but it is certain that it will have more public than medical appeal. From Page 3 (Chapter 1) in which some of the author's references are listed, namely Time, Life, and Readers Digest thru Page 219, one is given a resume of the pituitary-ovarian relationship with a discussion of pregnancy and of therapeutic considerations. One may rightfully wonder how a biochemist feels qualified to handle any clinical subjects as complicated as organotherapy. This speculation is confirmed by such startling (to the gynecologist) statements that "when acute pains and constant discomfort are associated with the onset of the menopause, some form of relief should be applied" (Page 150) or speaking of surgically castrate patients (Page 144), "A lemon color will appear in her face, indicative of the operations. Wrinkles will also make their appearance".

Actually this text is not without merit, for it covers an interesting if confused field in a thorough fashion. Of possible interest to the lay reader are the frequent expeditions into the fields of history, metaphysics, folk-lore, physiology, clinical and experimental medicine, but from the standpoint of any reviewing physician, this creates a veritable hodgepodge of information. Much of the material is based on half-truths and hypotheses that are medically uncertain and open to question, but are accepted and incorporated with dogma. This seems regrettable, but is hardly surprising when the author has had no clinical experience and has to rely on the many equivocal articles that permeate medical literature. Typical in this respect is the implicit faith in the excellent therapeutic results of the pituitary substances; which most practicing gynecologists would question.

This reviewer is generally rather tolerant and would like to close by emphasizing that the lay woman may enjoy a not too lengthy discussion of the function and relationship of her "female organs". She may be bored by such tedious descriptions of the collection of pregnant mare hormone (Pages 86 to 91), and she will be appalled at the things that are going to happen between the ages of 45 to 50 (Page 98). If she is not, however, too suggestible, she may derive considerable enjoyment from the text, and for that matter, any doctor may relish it as light reading, if he does not accept the context as clinically so definite as the author implies.

EDMUND R. NOVAK

BOOKS RECEIVED FOR REVIEW

Administrative Medicine. Transactions of the Second Conference December 8, 9 and 10, 1953. Edited by GEORGE S. STEVENSON. 164 pp., $3.00. The Josiah Macy, Jr. Foundation, New York, N. Y.

Biochemical Investigations in Diagnosis and Treatment. By JOHN D. N. NABARRO. 299 pp., 25s. net. H. K. Lewis & Co., London, England.

Coronary Heart Disease in Young Adults. By MENARD M. GERTLER AND PAUL D. WHITE. 218 pp., $5.00. Harvard University Press, Cambridge, Mass.

Essentials of Pediatrics, 5th edition. By JEANS, WRIGHT AND BLAKE. 808 pp., $4.75. J. B. Lippincott Co., Philadelphia, Pa.

Intracranial Gliomata. By JOHN PENMAN AND MARION C. SMITH. Medical Research Council

Special Report Series No. 284. 70 pp., $1.25. Her Majesty's Stationery Office, London, England.

Man: Mind Or Matter? By CHARLES MAYER. Translated and with a preface by HAROLD A. LARRABEE. 168 pp., $2.50. The Beacon Press, Boston, Mass.

Professional Adjustments, 3rd edition. By SISTER MARY ISIDORE LENNON. 381 pp., $4.75. The C. V. Mosby Co., St. Louis, Missouri.

The Study of the Brain. A Companion Text to the Stereoscopic Atlas of Neuroanatomy. By HYMAN S. RUBINSTEIN. Foreword by Carl L. Davis. 222 pp., $9.50. Grune & Stratton, New York, N. Y.

Symposium on Problems of Gerontology. Proceedings of a Symposium held under the auspices of The Johns Hopkins University, School of Hygiene and Public Health and The National Vitamin Foundation, New York City, March 2, 1954. By F. H. BETHELL, B. F. CHOW, C. S. DAVIDSON, H. J. DEUEL, JR., D. J. INGLE, J. E. KIRK, A. I. LANSING, C. N. H. LONG, H. A. RAFSKY AND N. W. SHOCK. 141 pp., $2.50. The National Vitamin Foundation, New York, N. Y.

Textbook of Bacteriology, 3rd edition. By JOSEPH M. DOUGHERTY AND ANTHONY J. LAMBERTI. 598 pp., $8.25. The C. V. Mosby Co., St. Louis, Missouri.

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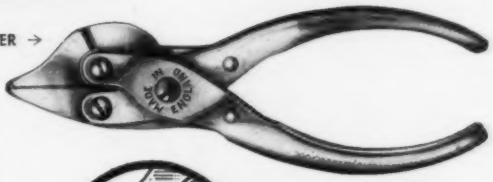

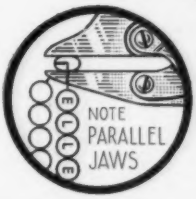

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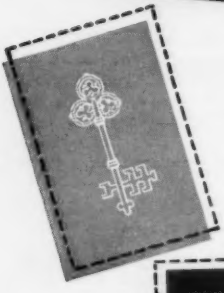

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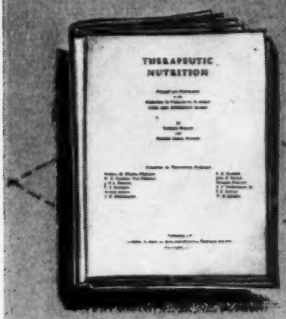

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